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# INHERITANCE STUDIES ON DURATION OF DEVELOPMENTAL STAGES IN CROSSES WITHIN THE GENUS LYCOPERSICON [1]

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## INTRODUCTION

This study deals with the inheritance of duration of three stages of development in certain crosses involving varieties of *Lycopersicon esculentum* Mill. and *L. pimpinellifolium* (Jusl.) Mill. These three stages are (1) number of days from seeding to first bloom, (2) number of days from first bloom to first fruit set, and (3) number of days from first fruit set to first complete change of color of any fruit. The sum of these three stages obviously represents the number of days from seeding to first ripe fruit and may be regarded as the measure of an earliness-of-maturity character.

The purposes of the investigation were to determine whether these several natural biological periods in the development of the tomato plant are definite subcharacters; to obtain information concerning heterosis and dominance; and to ascertain the efficiency of the fit between obtained and theoretical means, based on certain formulas (*2*, *24*, *29*),[2] as a method of determining whether the effects of the genes differentiating the quantitative characters are arithmetically or geometrically cumulative. The investigation was carried out at the Cheyenne Horticultural Field Station, Cheyenne, Wyo.

Throughout the study and the interpretation of the data, the authors have attempted to keep in mind the probability that the genes bring about the differentiation of a character by initiating (either directly or indirectly) developmental processes which, no doubt, in many cases interact among themselves (*10*, *11*, *15*, *20*, *29*, *32*, *42*, *43*).

## EXPERIMENTAL CONSIDERATIONS

The design of an experiment has considerable bearing upon the interpretation of the data. Such being the case, the purpose or purposes of an experiment play a considerable part in determining the experimental design. The design for the present study was based on genetic and statistical considerations. The genetic aspects will be considered first.

From the immediately foregoing statements, it is apparent that the experimental design should allow for testing whether the stages (number of days from planting to first bloom, number of days from first bloom to first fruit set, and number of days from first fruit set to

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[2] Italic numbers in parentheses refer to Literature Cited, p. 146.

first complete change of color of any fruit) are definite subcharacters, i. e., biologically sound subdivisions, of number of days from planting to first complete change of color of any fruit. To test the logic of such a division it is necessary to have parents that differ phenotypically as regards these stages. *Lycopersicon pimpinellifolium*, variety Red Currant, and *L. esculentum*, varieties Johannisfeuer, Danmark, and Bonny Best, were believed to have this diversity and for that reason were used in the study.

Next, the experiment should provide for measuring heterosis and dominance. Since heterosis and dominance may be determined by comparing the expression of a character in the $F_1$ with the expression of the same character in each of the parents, at least both parents and the $F_1$ should be included in the study.

Also, the design should allow for determining the efficiency of the method involving the fit between obtained and theoretical means for testing whether the effects of the genes are arithmetically or geometrically cumulative. Lindstrom (*21*), Powers (*29*), and Lyon (*23*) have pointed out the necessity, in such studies, of taking dominance into consideration. Therefore, in order to obtain the most information possible from any particular cross this method requires that both parents, the $F_1$ and $F_2$ generations, and the progenies obtained from backcrossing the $F_1$ to each parent be included.

One other genetic consideration in the design of the experiment should be mentioned. Since the facilities of most investigators are limited, one usually has the choice, in field investigations, between a larger sample of plant material, as represented by number of crosses studied, and a larger sample of environmental conditions, as represented by years. To obtain as large a sample of plant material as possible it would be necessary to grow different crosses every year; whereas to obtain as large a sample of years as possible, it would be necessary to grow the same crosses every year. In this study the authors were more interested in obtaining as large a sample of plant material as possible but did not want to neglect entirely environmental effects as represented by years. Consequently, the cross Johannisfeuer × Red Currant was grown in 1937 and 1939, the cross Johannisfeuer × Danmark was grown in 1938 and 1939, the cross Danmark × Red Currant was grown in 1938, and the cross Johannisfeuer × Bonny Best was grown in 1939. Furthermore, this design provided that Johannisfeuer, Red Currant, and Danmark be crossed in all combinations and that Johannisfeuer be crossed with all varieties used in the study. The sample of plant material was sufficient to allow the drawing of definite conclusions, and the sample of years was sufficient to furnish information on the interaction between environment and the genes differentiating the duration of stages of development.

The experimental design as regards the greenhouse and field technique was very similar to that reported by Powers (*28*, *29*). Consequently, only such details as are essential to an interpretation of the data will be given here. The parents and generations were grown in a randomized-block experiment, there being a total of 9 blocks in 1937 (with the exception of the $F_1$ generation, which will be mentioned later) and a total of 20 blocks in 1938 and again in 1939. One of the replications of the Bonny Best parent was lost in 1939. Yates' method (*44*) was used in calculating the missing plot. The material was randomized within blocks by the use of Tippett's tables (*38*), and the random-

ization was the same for the greenhouse and the field. The seed was sown on April 12 in 1937, on April 20, 21, and 26 in 1938, and on April 17 in 1939. The plants were spotted out in flats on April 26 and 27 in 1937, on May 5, 6, and 9 in 1938, and on May 1 and 2 in 1939. The material was transplanted into the field on May 26 and 27 in 1937, on June 1 in 1938, and on May 31 and June 2 in 1939. All operations were performed on the basis of blocks. Randomization within blocks prevented the danger of confounding any differences due to operations with differences due to crosses, parents, or generations.

The plots were 24 plants long in 1937 and 1938, and 12 plants long in 1939. In 1937 the plants were spaced 3½ feet apart within the plot and the plots were 3½ feet apart. This provided a spacing of 3½ by 3½ feet for plants. In both 1938 and 1939 the spacing was 4 by 4 feet. The greater spacing of 1938 and 1939 materially facilitated the taking of notes, as all notes were taken on the basis of individual plants.

The seeds were sown late in an effort to avoid any blooming of plants before transplanting into the field. In 1937 only two plants had bloomed before transplanting, in 1938 none, and in 1939 five. The material was grown under irrigation; and sufficient water was supplied during the spring, summer, and early fall to keep the plants in a vigorous growing condition.

In 1937, the $F_1$ generation was grown with the parents in a separate randomized-block planting because of the smaller number of $F_1$ plants available. All comparisons involving this generation are made through the parents in the two sets of randomized blocks. This accounts for the large standard errors of the $F_1$ and of the theoretical means for 1937.

One of the environmental influences should be mentioned. Severe hailstorms on July 15, 1937, and on July 13, 1938, caused quite uniform damage over the plantings in both years. To overcome possible differences due to hail injury, all plants were uniformly pruned after the hail, all remaining fruits, flowers, and flower buds being removed, In 1937 first-bloom data were obtained both before and after hail damage. Although, in 1937, 12 percent of the plants had not bloomed before the hail, the authors believe that these data taken before hail have value. Therefore, 2 days were added to the period from planting the seed to the date of the hailstorm, and this value was recorded as the number of days from the planting of the seed to first bloom for those plants that had not bloomed. This will be mentioned again in the interpretation of the results.

A clear understanding of the stages involved in the earliness-of-maturity character, as measured by number of days from seeding to first complete change of color of any fruit, is essential to a proper interpretation of the results. As previously stated, this character was divided into the following stages: Number of days from seeding to first bloom, number of days from first bloom to first fruit set, and number of days from first fruit set to first complete change of color of any fruit.

The individual plants were easily classified on the basis of these stages of development. In classifying plants for the first stage of development, the plants were recorded as having bloomed on the day that the petals opened out. For the second stage of development, the fruit was considered as having set when the ovary showed definite enlargement. The plants were examined regularly thereafter until

the fruits were definitely established on the plants, to determine if sloughing occurred. Under the conditions of this experiment, sloughing of fruits rarely if ever occurred. This second stage comprised the period after flowering necessary for definite enlargement of the ovary and the period during which flowers might have been sloughed. The period from the time of blooming of any one flower until its ovary has definitely enlarged is longer for some varieties than for others. Also, in some years, all of the blossoms do not set fruit, but instead an abscission layer is formed and the blossoms drop. Hence, the stage from first bloom to first fruit set is prolonged. In establishing the third stage of development, the first complete change of color was recorded as that time when the green in the fruit had disappeared. On any specified fruit this date could be determined within a day.

The method employed in reducing the data was essentially the same as that used by Powers (*29*), with the exception of some minor changes desirable because of the nature of the experiment. Only these changes need be mentioned here. Since the primary interest lay in comparisons between averages of parents or generations, the plot instead of the plant was used as the unit for reducing the data. Then, for each parent and generation the standard errors were estimated for within crosses from the averages of plots, which were derived from single-plant data. In all tables the standard errors of the means are given. Differences having $P$ values less than 0.05 were considered as statistically significant. The symbol $B_1$, which appears both in the tables and in the text, signifies that the progeny studied resulted from backcrossing the $F_1$ to the designated parent.

In either breeding or genetic studies the problems involved are generally simplified if the characters under investigation can be properly partitioned into their components. This is readily appreciated when one stops to consider that the number of genes differentiating any particular component of a character must be less than the number differentiating the character itself. Hence, both the breeding and genetic facts are made more intelligible by partitioning the character into its components. The earliness character, measured by the number of days from seeding to first complete change of color, with which these studies are concerned, seems to be composed of the three stages of development already specified; namely, number of days from seeding to first bloom, number of days from first bloom to first fruit set, and number of days from first fruit set to first complete change of color.

Before proceeding further, it is desirable to examine the data for the purpose of determining whether these stages of development are subcharacters of number of days from seeding to first complete change of color. If they are subcharacters, variation as to the duration of the different stages of development may be expected among the parents and generations. Likewise, different combinations of the duration of the different stages of development may be expected to occur among the variants (parents and generations). For example, all the stages of development may be of short duration in one variant, all of long duration in another variant, and the first two may be of short duration and the last of long duration in a third variant. Obviously, other combinations are possible but need not be given, as those already presented are sufficient to illustrate the point.

## EXPERIMENTAL RESULTS

### SUBDIVISIONS OF EARLINESS CHARACTER

#### JOHANNISFEUER × RED CURRANT AND DANMARK × JOHANNISFEUER

For the cross Johannisfeuer × Red Currant the data that have a bearing upon the problem as to whether the three stages of development form subcharacters of number of days from seeding to first complete change of color of any fruit are given in table 1. An examination of these data reveals that the period from seeding to first bloom is of short duration for Red Currant and the $F_1$ generation and of long duration for Johannisfeuer. For number of days from first bloom to first fruit set, the differences between means are not great. However, for 1939 the $F_1$ generation, in comparison with the parent varieties, has a definitely shorter period from first bloom to first fruit set. These same comparisons for 1937 are not conclusive because of the large standard error for the mean of the $F_1$ generation. The period from first fruit set to first complete change of color of any fruit is short for the $F_1$ generation and for Johannisfeuer. It is short for Red Currant also in 1937 but somewhat longer in 1939. The fact that the means of the $F_1$ generation are the smallest for the character number of days from seeding to first complete change of color of any fruit indicates that all three of the stages of development for the $F_1$ generation are of short duration.

For the cross Danmark × Johannisfeuer, the means, expressed in number of days (table 1), show that the period from seeding to first bloom is comparatively short for Danmark and comparatively long for Johannisfeuer. The means of the generations for this stage of development fall in between the means of the two parents, grading from one parent to the other, depending on the closeness of the genetic relationship. For the second stage of development, the $F_1$ generation has the smallest mean in each year and the means of the other generations approach the means of one or the other parent as the generation being considered approaches a particular parent in genetic relationship. There are no statistically significant differences between the means of the parents as regards number of days from first bloom to first fruit set. Danmark has a long period from first fruit set to first complete change of color of any fruit, whereas this same period for Johannisfeuer is short. The means of the different generations fall between the means of the two parents, becoming larger as the generations approach the Danmark parent in genetic relationship.

TABLE 1.— *Means, expressed in number of days, for different stages of development for the crosses Johannisfeuer × Red Currant in 1937 and 1939 and Danmark × Johannisfeuer in 1938–39*

JOHANNISFEUER × RED CURRANT

| Parent or generation | Period from seeding to first bloom in— | | Period from first bloom to first fruit set in— | | Period from first fruit set to first complete change of color in— | | Period from seeding to first complete change of color in— | |
|---|---|---|---|---|---|---|---|---|
| | 1937 | 1939 | 1937 | 1939 | 1937 | 1939 | 1937 | 1939 |
| | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* |
| Red Currant | 107.1±0.109 | 71.8±0.728 | 3.1±0.014 | 5.0±0.156 | 42.5±0.380 | 49.3±0.501 | 152.7±0.395 | 126.0±0.612 |
| $B_1$ to Red Currant | 106.9± .091 | 71.0± .763 | 3.1± .019 | 4.9± .153 | 41.8± .308 | 47.1± .535 | 151.8± .319 | 123.1± .726 |
| $F_1$ generation | 107.3± .540 | 71.5± .553 | 3.3± .389 | 3.9± .110 | 39.3±1.485 | 43.5± .472 | 149.9±1.111 | 118.9± .568 |
| $F_2$ generation | 108.1± .058 | 76.7± .920 | 3.2± .017 | 4.3± .142 | 42.0± .148 | 44.5± .608 | 153.3± .155 | 125.5± .644 |
| $B_1$ to Johannisfeuer | 108.4± .077 | 76.5± .586 | 3.3± .024 | 4.0± .125 | 41.4± .159 | 44.0± .498 | 153.2± .187 | 124.7± .950 |
| Johannisfeuer | 111.9± .271 | 86.4± .776 | 3.9± .076 | 4.5± .160 | 42.7± .238 | 45.2± .483 | 158.5± .425 | 136.1± .838 |

DANMARK × JOHANNISFEUER

| Parent or generation | Period from seeding to first bloom in— | | Period from first bloom to first fruit set in— | | Period from first fruit set to first complete change of color in— | | Period from seeding to first complete change of color in— | |
|---|---|---|---|---|---|---|---|---|
| | 1938 | 1939 | 1938 | 1939 | 1938 | 1939 | 1938 | 1939 |
| | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* |
| Johannisfeuer | 104.6±0.545 | 86.4±0.776 | 5.6±0.093 | 4.5±0.160 | 54.7±0.504 | 45.2±0.483 | 164.9±0.900 | 136.1±0.838 |
| $B_1$ to Johannisfeuer | 102.7± .318 | 81.6± .817 | 5.5± .064 | 4.0± .137 | 56.8± .372 | 47.6± .564 | 165.0± .463 | 133.3± .787 |
| $F_1$ generation | 101.1± .540 | 80.5± .681 | 5.1± .087 | 3.9± .119 | 59.5± .450 | 48.0± .516 | 165.6± .617 | 132.4± .440 |
| $F_2$ generation | 101.4± .283 | 80.6± .610 | 5.3± .064 | 4.4± .240 | 59.7± .282 | 49.9± .412 | 166.4± .437 | 134.9± .727 |
| $B_1$ to Danmark | 100.6± .303 | 80.2± .743 | 5.4± .051 | 4.4± .225 | 61.6± .318 | 50.3± .621 | 167.6± .382 | 134.8± .983 |
| Danmark | 99.4± .351 | 78.2± .829 | 5.7± .114 | 4.9± .257 | 65.0± .473 | 54.6± .563 | 170.0± .489 | 137.7± .632 |

### Danmark × Red Currant and Johannisfeuer × Bonny Best

The means, expressed in number of days, for different stages of development for the crosses Danmark × Red Currant and Johannisfeuer × Bonny Best are listed in table 2. As regards the Danmark × Red Currant cross, the $F_1$ generation has the shortest period for all three stages of development. Red Currant (tables 1 and 2) has a comparatively short period for all three stages of development, whereas Danmark (tables 1 and 2) has a comparatively short period from seeding to first bloom and a comparatively long period from first fruit set to first complete change of color of any fruit. From table 2 it can be seen that Bonny Best has a comparatively long period for all three stages of development, whereas Johannisfeuer has a long period from seeding to first bloom. The $F_1$ of Johannisfeuer × Bonny Best has a short period from seeding to first bloom and from first bloom to first fruit set. The period from first fruit set to first complete change of color of any fruit for the $F_1$ is only 2.6 days longer than the same period for Johannisfeuer.

Table 2.—*Means, expressed in number of days, for different stages of development for the crosses Danmark × Red Currant in 1938 and Johannisfeuer × Bonny Best in 1939*

| Parent or generation | Period from seeding to first bloom | Period from first bloom to first fruit set | Period from first fruit set to first complete change of color of any fruit | Period from seeding to first complete change of color of any fruit |
|---|---|---|---|---|
| DANMARK × RED CURRANT (1938) | | | | |
| | *Days* | *Days* | *Days* | *Days* |
| Red Currant | 98. 6±0. 352 | 5. 4±0. 128 | 53. 0±0. 564 | 156. 9±0. 822 |
| $B_1$ to Red Currant | 97. 9± . 310 | 5. 1± . 056 | 52. 0± . 428 | 155. 0± . 472 |
| $F_1$ generation | 97. 5± . 328 | 4. 8± . 064 | 51. 5± . 609 | 153. 8± . 730 |
| $F_2$ generation | 98. 5± . 308 | 5. 2± . 057 | 53. 0± . 312 | 156. 6± . 480 |
| $B_1$ to Danmark | 98. 3± . 302 | 5. 1± . 055 | 56. 4± . 257 | 159. 7± . 315 |
| Danmark | 100. 0± . 462 | 5. 6± . 103 | 64. 3± . 435 | 169. 8± . 544 |
| JOHANNISFEUER × BONNY BEST (1939) | | | | |
| Johannisfeuer | 86. 4±0. 776 | 4. 5±0. 160 | 45. 2±0. 483 | 136. 1±0. 838 |
| $B_1$ to Johannisfeuer | 84. 7± . 613 | 4. 7± . 166 | 46. 8± . 489 | 136. 1± . 765 |
| $F_1$ generation | 82. 9± . 776 | 4. 3± . 176 | 47. 8± . 479 | 134. 9± . 736 |
| $F_2$ generation | 83. 5± . 744 | 4. 5± . 170 | 49. 5± . 558 | 137. 5± . 851 |
| $B_1$ to Bonny Best | 84. 7± . 816 | 4. 5± . 155 | 50. 1± . 659 | 139. 2±1. 006 |
| Bonny Best | 86. 2± . 576 | 5. 2± . 222 | 55. 5± . 783 | 146. 8± . 908 |

### Summary for All Crosses

It is desirable at this time to summarize the evidence as to whether the designated stages of development are subcharacters of number of days from seeding to first complete change of color of any fruit. From the fact that the maximum difference between the means of any of the comparable variants was only 1.1 days for number of days from first bloom to first fruit set and from the fact that the largest mean for any variant was only 5.7 days, it may be concluded that this stage of development is of comparatively short duration for all the variants. However, statistically significant differences were shown to occur. All three stages of development were found to be of short duration for the $F_1$ generation of the cross Danmark×Red Currant. Red Currant was found to have short periods from seeding to first bloom and from first

fruit set to complete change of color, whereas Bonny Best was found to have long periods for these same two stages of development and in addition to have the longest period from first bloom to first fruit set of any variety or generation with which it is comparable. In Johannisfeuer a long period from seeding to first bloom is combined with a short period from first fruit set to first complete change of color of any fruit; whereas in Danmark a short period from seeding to first bloom is combined with a long period from first fruit set to first complete change of color. It is apparent that variations occur in the duration of these three stages of development. Also, the findings for Johannisfeuer and Danmark prove that these variations may occur in different combinations. Hence, one may conclude that number of days from seeding to first complete change of color of any fruit is composed of the three substages indicated and that therefore the divisions made are biologically sound.

It might be well to point out that the authors do not contend that the character studied might not be partitioned further to advantage. For example, it might be advantageous to divide the stage number of days from seeding to first bloom into number of days required for germination and number of days from germination to first bloom. This was not done because of the difficulties involved in obtaining individual plant data on germination.

## EFFECT OF ENVIRONMENT ON STAGES OF DEVELOPMENT

Having found that the stages of development as designated are biologically sound, it is desirable to examine the data for the purpose of determining the influence that the environment has upon an interpretation of the data. Information concerning the influence of environment may be obtained by comparing the number of days from seeding to first bloom before and after the hailstorm in 1937 and by comparing the differences between years for the crosses Johannisfeuer×Red Currant and Danmark×Johannisfeuer.

When interpreting the data for mean number of days from seeding to first bloom before and after hail for each parent and generation (table 3), it should be kept in mind that 12 percent of the plants had not bloomed before the hailstorm occurred and that, in calculating the means and standard errors, all of these plants were considered as having bloomed on what would have been the next date for taking bloom data. Bloom data were taken three times a week. It is apparent that the means of all the parents and generations would not be affected to the same degree by such a procedure but that the means of the later maturing parent and generations would be somewhat smaller than they should be in comparison with the means of the earlier maturing parent and generations. To take a specific example, the difference before hail of 6.9 days between the mean of Johannisfeuer and the mean of Red Currant is in all probability somewhat less than it would have been if hail had not occurred. With these facts in mind, the data listed in table 3 may be interpreted more logically. It can be seen that the rank as to magnitude before hail and after hail is the same for the parents and generations with one exception; that is, in the comparison of the mean of Red Currant with the mean of the $B_1$ to Red Currant. Since in no instance are the differences between the means of the Red Currant parent and the means of the progeny obtained by backcrossing the $F_1$ generation to Red Currant statisti-

cally significant, the exception noted cannot be attributed logically to an interaction between parents or generations and the environment but can be accounted for by chance fluctuation. The difference between the mean of Red Currant and the mean of Johannisfeuer before hail (6.9) is too much greater than the difference between means of the same varieties after hail (4.8) to be accounted for by the probable errors of random sampling. The same conclusion holds for other similar comparisons involving generations or generations and parents. Then the only apparent effect of hail damage has been to decrease the magnitude of the differences between parents, of differences between generations, and of differences between parents and generations. Such being the case, the interpretation of the data as to the biological justification of the division number of days from seeding to first bloom has not been affected by environmental conditions resulting from hail damage.

TABLE 3.—*Mean number of days from seeding to first bloom before and after hail for each parent and generation of the cross Johannisfeuer × Red Currant in 1937*

| Parent or generation | Period from seeding to first bloom | |
|---|---|---|
| | Before hail | After hail |
| | *Days* | *Days* |
| Red Currant | 84.2±0.277 | 107.1±0.109 |
| $B_1$ to Red Currrant | 84.7± .313 | 106.9± .091 |
| $F_1$ generation | 85.4±1.358 | 107.3± .540 |
| $F_2$ generation | 87.2± .129 | 108.1± .058 |
| $B_1$ to Johannisfeuer | 88.6± .114 | 108.4± .077 |
| Johannisfeuer | 91.1± .223 | 111.9± .271 |

In regard to the effect of environment, as represented by years, upon an interpretation of the data, the means for the cross Johannisfeuer × Red Currant for the years 1937 and 1939 (table 1) show that the rank of the stage number of days from seeding to first bloom is the same (within the limits of sampling error) for both years. Consequently, the same interpretation as to the biological soundness of this stage as a division of the character number of days from seeding to first complete change of color in any fruit would have been placed upon the data if the experiment had been conducted for only 1 of the 2 years.

As to the second stage, in 1937 the mean for the Red Currant variety was significantly smaller than the mean for Johannisfeuer, whereas in 1939 the reverse was true. The data pertaining to the second stage show that in 1938 the mean for Johannisfeuer was $5.6 \pm 0.093$ (table 1) and for Red Currant $5.4 \pm 0.128$ days (table 2). These means are not significantly different if odds as great as 19 : 1 against the difference noted being due to the probable errors of random sampling are accepted as a criterion of significance. The data do not agree for any of the 3 years. It would seem that environmental conditions as represented by years interact with parents and generations in such a manner that the ranking of the means of the parents is not the same for the different years. The environment must be taken into account in studying this stage of development. However, the conclusions as to the soundness of considering this stage

of development a subdivision of the character under investigation is shown by the data for both 1937 and 1939.

Finally, in regard to the third stage of development for the cross Johannisfeuer×Red Currant (table 1) a detailed discussion of the data is not necessary, as it is apparent that in general the conclusions are the same as those drawn for the stage number of days from first bloom to first fruit set.

The data for the cross Danmark×Johannisfeuer in 1938–39 (table 1) show that for all three stages the rank of the means as to magnitude is the same for both years, with the possible exception of comparisons involving the $B_1$ to Johannisfeuer, the $F_2$ generation, and the $B_1$ to Danmark. These possible exceptions are for number of days from first bloom to first fruit set. Even these exceptions cannot be attributed to interactions between the environment and generations, as the differences between generations can be accounted for by the probable errors of random sampling (*39, p. 54, table 10*). In these cases, also, the interpretation placed upon the data as to the biological soundness of the stages of development as subdivisions of the character number of days from seeding to first complete change of color of any fruit is the same for both 1938 and 1939.

In summarizing the data for both crosses, it can be said that the environmental conditions as represented by years do not alter the interpretation placed upon the data as to whether the designated stages of development are justified biologically. This does not mean that environmental conditions do not have an influence upon the duration of the different stages of development. On the contrary, the data in table 1 lead one to conclude that environment has a decided effect on all stages of development.

## HETEROSIS AND DOMINANCE

Accepting Jones' well-known theory (*16*) of heterosis as being the most satisfactory for explaining the known facts regarding this phenomenon, it seems logical to discuss heterosis and dominance under the same heading. This does not mean that the authors consider that, from the standpoint of physiological genetics, heterosis and dominance are fundamentally different. In fact Jones' hypothesis (*16*) for the explanation of heterosis assumes at least the existence of partial dominance of the factors affecting the character under consideration. In this study the term "heterosis" is applied to those cases in which the mean period of development of the $F_1$ is less than the mean of either parent. In other words, the ability to complete the different stages of development in a shorter period of time is taken to be an expression of heterosis. The authors would like to point out that they do not mean to imply that a different criterion of heterosis would not be desirable for other studies.

Complete dominance is attributed in those instances in which the phenotype of the $F_1$ is that of one or the other parent. At the other extreme, when the phenotype of the $F_1$ generation is the arithmetic mean of the phenotypes of the two parents, dominance is considered to be nonexistent. Finally, when the mean of the $F_1$ generation falls between the means of the two parents but not exactly midway (arithmetically), the parental phenotype most closely resembled is considered to be partially dominant. Hence, like Mendel (*25*), Naudin (*26*), and other earlier workers, the authors have considered dominance

to be a phenomenon of the phenotype. Also, since the means of those $F_1$'s exhibiting heterosis fit neither the arithmetic nor the geometric mean calculated from the means of the two parents, it seems desirable at this time to use the earlier method of determining dominance; namely, that of ascertaining which parental phenotype the $F_1$ more nearly approaches.

### Johannisfeuer × Red Currant

The data for the cross Johannisfeuer × Red Currant (table 1) show that significant differences between the means of Red Currant, the $B_1$ to Red Currant, and the $F_1$ generation do not exist for number of days from seeding to first bloom for either 1937 or 1939. For all practical purposes the phenotype of the Red Currant parent is completely dominant. Also, it can be seen that in 1939 a significant difference did not exist between the mean of the $F_2$ and the mean of the $B_1$ to Johannisfeuer for the same subcharacter. The difference for 1937, although statistically significant, is small. The means of the Johannisfeuer parent for 1937 and 1939 are significantly larger than the means for any other variant.

Considering the data for number of days from first bloom to first fruit set, it can be seen that for 1937 the standard error of the mean of the $F_1$ generation is too large to allow the drawing of conclusions regarding either heterosis or dominance. For 1939 it is clearly evident that heterosis does exist, as the $F_1$ mean is significantly smaller than the mean of either parent, and furthermore in no case is the mean of any of the generations as large as the mean of the parent to which the generation is most closely related. However, the differences noted in this latter comparison are not always statistically significant.

The data for the third stage likewise show that the mean of the $F_1$ is significantly smaller than the mean of either parent for each year of this test. This shows that heterosis does occur for this stage of development. Also, each of the generation means, with the exception of that of the $B_1$ to Red Currant in 1939, is smaller than the mean for either parent. Again, the differences noted in this latter comparison are not always statistically significant.

For the earliness character, measured by number of days from seeding to first complete change of color, the means of the $F_1$ generation are significantly smaller than the mean for either parent for both 1937 and 1939, and furthermore each of the generation means for 1939 is smaller than the mean for either parent, although the differences are not always statistically significant.

### Danmark × Johannisfeuer

The data for the cross Danmark × Johannisfeuer (table 1) show that the mean of the $F_1$ generation falls between the means of the two parents for number of days from seeding to first bloom but is closer to the mean of the Danmark parent. Hence, earliness of bloom is partially dominant. The means of all the generations fall between the means of the two parents. Considering both 1938 and 1939, the differences between generation and parental means are statistically significant.

Regarding number of days from first bloom to first fruit set, the mean of the $F_1$ generation for each year is significantly smaller than the mean of either parent. Again, for both years the means of all

generations are smaller than the mean for either parent. However, the differences are not always statistically significant.

For number of days from first fruit set to first complete change of color, the mean of the $F_1$ generation lies between the means of the two parents but closer to the mean of the parent having a short period for this stage of development, and each of the generation means lies between those of the two parents. By the criterion used, no heterosis exists; but as was found for the first stage, short duration of the period involved is partially dominant.

From an examination of the data for number of days from seeding to first complete change of color, which is the summation of the previous stages, it is found that in 1938 the means of Johannisfeuer, the $B_1$ to Johannisfeuer, and the $F_1$ are not significantly different, and that each of the generation means falls between the means of the two parents. This would indicate complete dominance of short duration of the period from seeding to first complete change of color of any fruit. For 1939 this same character showed heterosis, the period from seeding to first complete change of color of any fruit being significantly shorter for the $F_1$ than for either parent. The same was true for all other generations as well, but the differences between the generation means and the parental means were not always statistically significant.

### Danmark × Red Currant and Johannisfeuer × Bonny Best

The data concerning heterosis and dominance for the crosses Danmark × Red Currant and Johannisfeuer × Bonny Best are given in table 2. In the case of the cross Danmark × Red Currant, all subcharacters and the character number of days from seeding to first complete change of color of any fruit have $F_1$ means significantly smaller than the comparable means of either parent, with the exception of the second subcharacter, in which case the difference divided by the standard error gives a $P$ value somewhat larger than 0.05. Accordingly, with the possible exception of number of days from first fruit set to first complete change of color of any fruit, all the stages of development and the combination exhibit heterosis. In the cross Johannisfeuer × Bonny Best, heterosis is exhibited for the first and second stages. However, in the latter the difference between the $F_1$ and the Johannisfeuer parent is not statistically significant. For the first subcharacter, each of the generation means is less than the mean of either parent. For number of days from first fruit set to first complete change of color of any fruit, the mean of the $F_1$ generation lies between the means of the two parents but is closer to the mean of Johannisfeuer. Hence, the shorter period of this stage of development is partially dominant. The $F_1$ mean for number of days from seeding to first complete change of color of any fruit is smaller than that of either parent. However, the difference between the $F_1$ mean and the mean of the Johannisfeuer parent is not statistically significant.

### Summary for All Crosses

Heterosis occurred in the Danmark × Red Currant and the Johannisfeuer × Bonny Best crosses, as is indicated by smaller number of days from seeding to first bloom. On the basis of number of days from first bloom to first fruit set, heterosis occurred in all crosses in all years, with the possible exceptions of Johannisfeuer × Red Currant in

1937 and Johannisfeuer × Bonny Best in 1939. In the Johannisfeuer × Red Currant cross, because of the large standard error of the $F_1$ generation, no definite conclusions could be drawn. Heterosis for the third developmental stage was exhibited by the cross Johannisfeuer × Red Currant. For this same stage, the mean of the $F_1$ generation in the cross Danmark × Red Currant was smaller than the mean of either parent but the difference between the mean of this generation and the mean of the Red Currant parent was not statistically significant. The character number of days from seeding to first complete change of color of any fruit exhibited heterosis in the crosses Johannisfeuer × Red Currant, Danmark × Johannisfeuer (in 1939 but not in 1938), and Danmark × Red Currant. For this character, the mean of the $F_1$ in the cross Johannisfeuer × Bonny Best was less than the mean of either parent but the difference involved in the comparison with Johannisfeuer was not statistically significant. In conclusion, it can be said that both heterosis and dominance are dependent upon the cross and upon environment as represented by years.

### EFFICIENCY OF THEORETICAL MEANS IN DETERMINING WHETHER THE EFFECTS OF THE GENES ARE ARITHMETICALLY OR GEOMETRICALLY CUMULATIVE

MacArthur and Butler (*24*), Charles and Smith (*2*), and Powers (*29*) have used the fit between obtained and theoretical means as a method of determining whether the data could be explained more logically on the assumption that the effects of the genes are arithmetically cumulative or on the assumption that they are geometrically cumulative. Much of the theory basic to the validity of this method is given by Charles and Smith (*2*) in developing the formulas for obtaining the theoretical means. This theory need not be repeated here. The necessity of considering dominance of the characters with which they worked was pointed out by Lindstrom (*21*) and Powers (*29*). Table 4 shows the formulas for estimating the theoretical means that take dominance and heterosis into consideration.

The formulas for the means of the $F_2$ generation were developed by Wright, as cited by Powers (*29*). In applying these formulas, in other words, in using this method, one should ascertain the efficiency of the method. If it is efficient, the difference between the two comparable theoretical means of generation or backcross must be statistically significant or, expressed in another way, the contrasted formulas must be discriminatory. If they are not discriminatory, the difficulty may be inherent in the data or it may be inherent in the method as applied to such data.

TABLE 4.—*Formulas for estimating arithmetic and geometric means*

| Generation or backcross | Arithmetic mean | Geometric mean |
|---|---|---|
| $F_2$ generation | $\frac{\overline{P}_1+2\overline{F}_1+\overline{P}_2}{4}$ | Antilogarithm of $\frac{\log \overline{P}_1+2\log \overline{F}_1+\log \overline{P}_2}{4}$ |
| $B_1$ to $P_1$ | $\frac{\overline{F}_1+\overline{P}_1}{2}$ | Antilogarithm of $\frac{\log \overline{F}_1+\log \overline{P}_1}{2}$ |
| $B_1$ to $P_2$ | $\frac{\overline{F}_1+\overline{F}_2}{2}$ | Antilogarithm of $\frac{\log \overline{F}_1+\log \overline{P}_2}{2}$ |

The data may be tested by two criteria: (1) If the differences between parents are of sufficient magnitude to be of economic importance, methods should be developed by which problems involving these differences can be studied; (2) if the differences are of sufficient magnitude to be established as statistically significant, methods should be developed by which problems involving these differences can be studied because such differences are important to theoretical genetics if not at the moment of importance to applied genetics. From tables 1 and 2 it can be seen that, with the possible exception of the stage from first bloom to first fruit set, economically important differences exist for all stages of development and for the earliness character measured by number of days from seeding to first complete change of color of any fruit. Hence any difficulties noted would not be inherent in the data.

TABLE 5.—*Obtained and theoretical means for different stages of development of the cross Johannisfeuer × Red Currant, expressed in number of days*

| Year and parent or generation | Period from seeding to first bloom | | | Period from first bloom to first fruit set | | |
|---|---|---|---|---|---|---|
| | Obtained | Theoretical | | Obtained | Theoretical | |
| | | Arithmetic | Geometric | | Arithmetic | Geometric |
| *1937* | | | | | | |
| | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* |
| $B_1$ to Red Currant | 106.9±0.091 | 107.2±0.275 | 107.2 | 3.1±0.019 | 3.2±0.195 | 3.2 |
| $F_2$ generation | 108.1± .058 | 108.4± .280 | 108.4 | 3.2± .017 | 3.4± .195 | 3.4 |
| $B_1$ to Johannisfeuer | 108.4± .077 | 109.6± .302 | 109.6 | 3.3± .024 | 3.6± .198 | 3.6 |
| *1939* | | | | | | |
| $B_1$ to Red Currant | 71.0± .763 | 71.7± .457 | 71.7 | 4.9± .153 | 4.5± .095 | 4.4 |
| $F_2$ generation | 76.7± .920 | 75.3± .384 | 75.0 | 4.3± .142 | 4.3± .078 | 4.3 |
| $B_1$ to Johannisfeuer | 76.5± .586 | 79.0± .476 | 78.6 | 4.0± .125 | 4.2± .097 | 4.2 |

| Year and parent or generation | Period from first fruit set to first complete change of color | | | Period from seeding to first complete change of color | | |
|---|---|---|---|---|---|---|
| | Obtained | Theoretical | | Obtained | Theoretical | |
| | | Arithmetic | Geometric | | Arithmetic | Geometric |
| *1937* | | | | | | |
| | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* |
| $B_1$ to Red Currant | 41.8±0.308 | 40.9±0.766 | 40.9 | 151.8±0.319 | 151.3±0.590 | 151.3 |
| $F_2$ generation | 42.0± .148 | 41.0± .751 | 40.9 | 153.3± .155 | 152.5± .574 | 152.7 |
| $B_1$ to Johannisfeuer | 41.4± .159 | 41.0± .752 | 41.0 | 153.2± .187 | 154.2± .595 | 154.1 |
| *1939* | | | | | | |
| $B_1$ to Red Currant | 47.1± .535 | 46.4± .344 | 46.3 | 123.1± .726 | 122.5± .417 | 122.4 |
| $F_2$ generation | 44.5± .608 | 45.4± .293 | 45.3 | 125.5± .644 | 125.0± .385 | 124.8 |
| $B_1$ to Johannisfeuer | 44.0± .498 | 44.4± .338 | 44.3 | 124.7± .950 | 127.5± .506 | 127.2 |

To determine the efficiency of the method as applied to these data, the obtained and theoretical means for different stages of development in the cross Johannisfeuer×Red Currant are tabulated in table 5. In no case is there a statistically significant difference between the obtained mean and the corresponding theoretical mean. The comparable data in the latter half of table 1 and in table 2 for the other crosses were calculated; but, since the conclusions drawn

were the same as those drawn for the data presented in table 5, it is not necessary to present the data in detail. It is apparent that, as regards this study, this method is not sufficiently sensitive to be of any particular value as a means of studying the nature of the interactions of the genes. The agreement between the obtained means and the theoretical means was good with the possible exception of the $B_1$ to Johannisfeuer in 1939 for number of days from seeding to first bloom (table 5). Also, the agreement between the obtained and the theoretical means was good for the other crosses. Because of the inefficiency of the method employed, conclusions drawn from such facts would be of little, if any, value.

## DISCUSSION AND CONCLUSIONS

From the experimental data it was found that the earliness-of-maturity character, as measured by number of days from seeding to first complete change of color of any fruit, is composed of at least three stages of development: Number of days from seeding to first bloom, number of days from first bloom to first fruit set, and number of days from first fruit set to first complete change of color of any fruit. This fact has considerable importance in breeding early varieties of tomatoes. The cross Danmark × Johannisfeuer may be used to illustrate this point. It will be remembered that Danmark has a short period of development from planting of the seed to first bloom but a long period of development from first fruit set to first complete change of color of any fruit, whereas Johannisfeuer has a long period of development from planting of the seed to first bloom but a short period of development from first fruit set to first complete change of color of any fruit. Now, since the $F_1$ generations of the crosses Danmark × Red Currant and Johannisfeuer × Red Currant have short periods for both of these stages of development, it would seem that a variety could be produced recombining the genes for short duration of the stage of development from seeding to first bloom, characteristic of Danmark, with the genes for short duration of the stage of development from first fruit set to first complete change of color of any fruit, characteristic of Johannisfeuer. Adverse linkage relationships would complicate the problem. Also, the nature of the interactions of the recombined genes would determine whether a strain could be developed that possessed a short period for both of these stages of development. At the present time neither the linkage relationships nor the nature of the interactions of the genes are known.

The importance of heterosis to a breeding program was recognized by early workers: Kölreuter (*19*), Knight (*18*), Sageret (*31*), Gärtner (*9*), Focke (*8*), Shull (*33*, *34*, *35*, *36*), East (*4*, *5*, *6*), East and Hayes (*7*), Jones (*17*), and many others. The possibility of utilizing heterosis in commercial production of tomatoes has been pointed out by Wellington (*40*), Hayes and Jones (*14*), and others. Jones (*17*), reporting on earliness in tomatoes, a large part of the data having been collected by H. K. Hayes, states that in one cross the $F_1$ generation was fully as early as the early parent. Luckwill (*22*) found that the time of flowering in tomato hybrids was intermediate between that of their parents, except in intraspecific crosses that differed in a major growth factor (*d* or *br*), when dominance of early flowering was shown. In regard to utilizing heterosis in tomato production, the possibility of making profitable the use of $F_1$ seed to produce the commercial

crop of tomatoes should be considered. The authors realize that this problem cannot be completely solved at this time. However, the data on earliness of maturity as reported in this paper have some bearing upon the solution.

The data having a bearing upon heterosis as expressed by shortening of the period from seeding to first complete change of color of any fruit are listed in table 6. Upon examination of these data, it is found that only 2 of the 12 differences are not statistically significant. The $F_1$ generations are not significantly earlier than Johannisfeuer in either the cross Danmark × Johannisfeuer in 1938 or the cross Johannisfeuer × Bonny Best. Special attention should be given to the crosses Johannisfeuer × Red Currant and Danmark × Red Currant. For the cross Johannisfeuer × Red Currant, the period from seeding to first complete change of color in 1937 was 2.8 days shorter for the $F_1$ than for the Red Currant parent and 8.6 days shorter for the $F_1$ than for the Johannisfeuer parent; in 1939 this same period was 7.1 days shorter for the $F_1$ than for the Red Currant parent and 17.2 days shorter than for the Johannisfeuer parent. For the cross Danmark × Red Currant, the period from seeding to first complete change of color of any fruit was 16 days shorter for the $F_1$ than for Danmark and 3.1 days shorter than for Red Currant. It may be concluded that heterosis is quite pronounced in the cross Johannisfeuer × Red Currant and the cross Danmark × Red Currant.

TABLE 6.—*The differences obtained by subtracting the parental means from their respective $F_1$-generation means for number of days from seeding to first complete change of color of any fruit*

| Cross and parent | Differences[1] between means (cross minus parent) | | |
|---|---|---|---|
| | 1937 | 1938 | 1939 |
| | *Days* | *Days* | *Days* |
| Johannisfeuer × Red Currant: | | | |
| Johannisfeuer | −8.6±1.190 | | −17.2±1.012 |
| Red Currant | −2.8±1.179 | | −7.1± .835 |
| Danmark × Johannisfeuer: | | | |
| Danmark | | −4.4±0.787 | −5.3±0.770 |
| Johannisfeuer | | *.7±1.091 | −3.7± .946 |
| Danmark × Red Currant: | | | |
| Danmark | | −16.0± .910 | |
| Red Currant | | −3.1±1.099 | |
| Johannisfeuer × Bonny Best: | | | |
| Johannisfeuer | | | *−1.2±1.115 |
| Bonny Best | | | −11.9±1.169 |

[1] $P < 0.01$ for all differences not marked with an asterisk (*), but $> 0.05$ for all values marked with an asterisk.

In evaluating the shortened period from seeding to first complete change of color of any fruit exhibited by the $F_1$ generation, it is well to consider some of the medium- to large-fruited varieties available for commercial production. Babb and Kraus (*1*) have found Johannisfeuer to be the earliest in maturity of the medium- to large-fruited varieties and Danmark to be the earliest variety that produces fruits of sufficient size, smoothness, and quality to be classified as a fairly acceptable commercial variety. Bonny Best is classified as a second early by Boswell [3] and is one of the most widely grown varieties. In 1939 the $F_1$ of Johannisfeuer × Red Currant required an average of 17.2 fewer days from seeding to first complete change of color of any

[3] Personal correspondence.

fruit than did Johannisfeuer, and 27.9 fewer days than did Bonny Best. In 1938 the $F_1$ generation of Danmark × Red Currant required an average of 16.0 fewer days from seeding to first complete change of color of any fruit than did Danmark. These figures amply demonstrate the advantage in earliness that these $F_1$ generations have over the earliest maturing commercial varieties. If this advantage of the $F_1$ generations can be transferred to the $F_1$ generations of commercial varieties, such a new production will be of decided benefit to those sections having a short growing season; for example, the high irrigated valleys of the Rocky Mountains and the northern parts of the United States. It seems possible that the use of $F_1$ seed may have a place in commercial tomato production, the $F_1$ being grown until the plant breeder, if possible, recombines the desirable characteristics of both varieties into a single variety as early as the $F_1$.

The great importance to the plant breeder of information concerning the nature of the interactions of the genes affecting earliness of maturity may be deduced from the foregoing discussion. Therefore, methods of studying the nature of the interactions of genes differentiating quantitative characters merit careful consideration.

Three methods that have been used to study the nature of the interactions of the genes differentiating quantitative characters will be discussed. In some studies the genotypes can be identified directly (*13*, *27*, *41*), and thus segregates of different genotypes can be accurately classified. Where such a procedure can be followed, this is probably, in general, the most efficient method of studying the nature of the interactions of the genes. However, in the majority of cases, the different genotypes dependent upon segregation of the genes differentiating the quantitative character cannot be determined. Then the nature of the interactions of the genes may be studied by means of markers (*3*, *28*, *29*, *30*, *37*) and possibly by comparing the averages of different generations with a predicted average calculated from parental and $F_1$ means. The calculation of these means is based on certain theoretical considerations (*2*, *12*, *24*, *29*).

All three of these methods have certain advantages and if applicable should furnish information concerning the nature of the interactions of the genes as measured by end products. An advantage of the first method is that genes having major effects are being studied directly through the effects they produce. However, it should be kept in mind that other genes on the same chromosome that differentiate the same character would have an effect also. An advantage of the second method is the ease with which the classification of the segregating generations can be made. By a proper design of the experiment the effects of sections of the chromosome can be measured and the nature of the interactions of the genes studied (*30*). The third method entirely dispenses with the classification within generations. As a consequence, both the work of taking the data and the work of reducing it are greatly diminished, making it possible to broaden the scope of the studies by including more crosses. Certain other facts should be kept in mind regarding this method. Lindstrom (*21*), Powers (*29*), and Lyon (*23*) have shown that dominance must be taken into account also. This may be summed up by saying that the measurements of the $F_1$ must be used in calculating the theoretical means. Moreover, it should be kept in mind that the generations are treated as the units and hence it is not the nature of the interactions of

particular gene pairs that is measured but probably the nature of the interactions of a large number of gene pairs whose individual effects have not been determined. In other words, this type of study deals with the genes in mass. It is apparent that the information obtained by the use of one of these methods would supplement that obtained by the use of another. Hence, providing they are efficient, all would have their place in studying the nature of the interactions of the genes as measured by end products. It will be remembered that the method of comparing obtained parental and generation means with theoretical means was not sufficiently discriminative to be of any value in determining the nature of the interactions of the genes differentiating the stages of development under consideration in this study.

## SUMMARY

The subdivision of the period from seeding to first complete change of color of any fruit into the following stages of development was found to be biologically sound for certain crosses involving varieties of *Lycopersicon esculentum* and *L. pimpinellifolium:* (1) Number of days from seeding to first bloom; (2) number of days from first bloom to first fruit set; (3) number of days from first fruit set to first complete change of color of any fruit.

In each stage of development heterosis was exhibited in the case of some one or other of the crosses.

Heterosis was found to be dependent upon both the genotype (cross) and the environment.

The method involving comparison of the obtained mean of a given generation with predicted arithmetic and geometric means calculated from obtained parental and $F_1$ means was of little value, as regards these data, in determining whether the nature of the interactions of the genes was such that the effects were arithmetically cumulative or whether it was such that the effects were geometrically cumulative.

The possible importance of the findings in a plant-breeding program is discussed.

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# INHERITANCE OF QUANTITATIVE CHARACTERS IN CROSSES INVOLVING TWO SPECIES OF LYCOPERSICON [1]

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## INTRODUCTION

Studies on the inheritance of quantitative characters in crosses within the genus *Lycopersicon*, like all quantitative inheritance studies, are concerned with multiple factors, heterosis, dominance, and the nature of the interactions of the genes differentiating the quantitative characters. The purpose of the research reported herein was to obtain more information concerning the latter three phases of the problem and to apply this information in shaping a tomato-breeding program. The work was done at the Horticultural Field Station, Cheyenne, Wyo.

Before proceeding further, the writer wishes to point out that, even though from the standpoint of physiological genetics heterosis and dominance are probably fundamentally the same, it is convenient in these studies to use both terms. It is important, however, that the use of these terms be clearly understood. Those cases are considered as exhibiting heterosis in which the phenotypic expression of a character in the $F_1$ generation is either greater than or less than the magnitude of such expression in either parent. Complete dominance is applied to those cases in which the phenotypic expression in the $F_1$ generation is that of one or the other parent, provided there is a significant difference between the two parents as regards the character under consideration. The term "partial dominance" is used when the phenotypic expression in the $F_1$ generation lies between that of the parents, but is anything other than exactly intermediate (arithmetically). Finally, neither heterosis nor dominance is considered as existing when the expression in the $F_1$ generation is exactly intermediate (arithmetically) between the magnitudes for the two parents.

## PREVIOUS RESEARCH

Without attempting to review all of the previous investigations dealing with heterosis and dominance and with the nature of the interactions of the genes as measured by end products, it is desired to recall the fundamental principles established by earlier work, as well as some of the theories dealing with the genetic explanation of these phenomena.

### HETEROSIS AND DOMINANCE

Hybrid vigor has received the attention of plant students for more than a century and a half. Kölreuter (*29*) [2] reported cases of hybrid

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[2] Italic numbers in parentheses refer to Literature Cited, p. 172.

vigor as early as 1766. That hybrid vigor is of general occurrence in those $F_1$ generations that come from parents of genetic diversity was established early by Knight (*28*), Sageret (*41*), Gärtner (*13*), Darwin (*4*), and Focke (*12*). Inbreeding followed by hybridization, as a method of utilizing heterosis in corn improvement, was advocated by Shull (*42*, *43*, *44*), East (*6*, *7*, *8*), and East and Hayes (*11*). Their work was the basis for the development of the modern methods employed in breeding those crops in which the utilization of heterosis is a primary consideration.

The first genetic explanation of hybrid vigor was given by Keeble and Pellew (*25*) in 1911. They crossed two varieties of peas of medium size and obtained $F_1$ plants larger than either parent. One of the parents used in the cross had few but long internodes, whereas the other parent had numerous, short, thick internodes. The $F_1$ hybrid had both numerous and long internodes and therefore was considerably taller than either parent. Keeble and Pellew (*25*) found that each of these characters was differentiated by one factor pair and that the larger size of the $F_1$ generation was due to the fact that the effects of these two gene pairs were cumulative.

That there is an association between number of dominant factors and hybrid vigor was postulated by Bruce (*2*). Jones (*23*) followed up this supposition and developed what is probably the most generally accepted explanation of heterosis. This hypothesis is, briefly, as follows: (1) A large number of genes are responsible for the differentiation of most of the quantitative characters; and (2) those genes favorable to the production of a quantitative character are at least partially dominant. These two assumptions are sufficient to account for hybrid vigor, but may not explain why corn breeders have not been able to obtain selfed lines of corn as vigorous as the $F_1$ generation between two inbred lines that combine well. Jones (*23*) pointed out that, since maize has only 10 pairs of chromosomes, if a large number of gene pairs are involved some of these genes would have to be linked provided the chromosome theory of heredity is correct. Consequently, the probability would be extremely small of recombining into a single inbred line all of the genes favorable to growth of two inbred lines. Most plant breeders seeking to take advantage of heterosis have accepted this theory and have used it as a basis for their breeding programs. As is well known, corn breeders have been particularly successful in using breeding methods based on Jones' (*23*) explanation of heterosis (*18*, *27*).

In 1936 East (*10*) advanced a somewhat different theory in explanation of heterosis. He divided genes into two classes, defective and nondefective, and stated that heterosis is not concerned with the defective genes. With this conception in mind, East (*10*) made the following statement: "The cumulative action of the nondefective allelomorphs of a given gene approaches the strictly additive as they diverge from each other in function." With this classification of genes in mind, East (*10*) developed the following theory. If numerous nondefective allelomorphs are common in any given species, heterosis can be accounted for as follows: The effect of $A_1$ $A_1$ is not 2 $A_1$, but 2 $A_1-\alpha$, where the value of $\alpha$ approaches the value of $A_1$. But if $A_1$ . . . $A_4$ are a series of nondefective allelomorphs of $A_1$, wth their functions departing from those of $A_1$ on an ascending scale, then the resultant

effects may be visualized as $A_1\ A_2$ equaling $A_1+A_2-\beta$, $A_1\ A_3$ equaling $A_1+A_3-\gamma$, and $A_1\ A_4$ equaling $A_1+A_4-\delta$, where $\alpha>\beta>\gamma>\delta$.

At this time it is desirable to compare the two theories. Both assume that a large number of factors are involved in differentiating the quantitative characters. Jones' (*23*) theory assumes at least partial dominance of these numerous favorable growth genes, whereas East's (*10*) theory assumes divergence in function between members of any one allelomorphic series. To explain why corn breeders have not been able to select inbred lines as vigorous as the more productive $F_1$ generation hybrids, Jones (*23*) points out that if the genes bringing about heterosis are very numerous in a species, such as *Zea mays*, with only 10 pairs of chromosomes, some of the genes favorable to growth would of necessity be linked with genes not so favorable to growth. Consequently, it would be extremely difficult to obtain inbred lines as vigorous as the $F_1$ generation. According to East's (*10*) theory, if only divergence in function between members of pairs of allels were operating to bring about the expression of hybrid vigor, the production of inbred lines as vigorous as the $F_1$ generation, originating from genetically diverse parents, would be impossible.

The evidence is not very extensive in support of the contention that divergence in function occurs between members of allels. Sinnott and Dunn (*47*) cite a case reported by Dunn, in which each of two recessive genes ($t^0$ and $t^1$) in the mouse is lethal when homozygous ($t^0t^0$ or $t^1t^1$), but the heterozygote ($t^0t^1$) is viable and normal in character. They point out that the action of these two probably allelic genes is complementary and hence the results can be interpreted on the basis of divergence in function of the members of the allelic pair of genes. Luckwill (*32*) believes that the results he obtained from studies dealing with heterosis can be most logically explained by East's (*10*) theory. It is clear that the partial dominance theory for the explanation of heterosis and the theory involving the divergence in function of members within allels are not mutually exclusive, since the phenomena differentiating these two theories may be operating in the same cross. That such may be the case seems probable when one considers the part that duplication of parts of chromosomes, aneuploidy, and polyploidy accompanied by point mutation must have played in the origin of divergent plant forms. Hence, both of these theories merit careful study.

Evidence sufficient to differentiate the partial-dominance theory and the theory based on divergence in function between members of allels will be difficult to obtain. However, evidence concerning the relative importance of these two phenomena will not be so difficult to obtain. For example, if it were possible to establish selfed lines as vigorous as the $F_1$ generation, in those cases in which the $F_1$ generation was materially more vigorous than the inbred lines from which it originated, then the divergence in function of members of allels, at least, could not have played a predominant part in bringing about heterosis; since, according to this theory, heterosis is dependent upon heterozygosity. The experimental design for such a test should include at least the $F_1$ generation, the original inbred lines from which it sprang, and the selected inbred line or lines. As far as the author can ascertain, no case involving such a critical test in which the selected inbred lines have equaled the $F_1$ generation in vigor has been

reported for either naturally self-fertilized or naturally cross-fertilized plants.

Richey and Sprague (*40*) have done work that has a bearing upon this problem. They attempted to increase productivity of inbred lines by convergent improvement (*39*). The process involved backcrossing the $F_1$ generation to each of the original inbred lines entering the cross to produce the $F_1$ generation, and then selecting to carry over as many of the favorable factors from the nonrecurring parent as possible. These selected plants were again crossed to the recurring parent. Hence, two independent backcrossing programs were carried along simultaneously, one to one parent and another to the other. The criterion as to the number of times that backcrossing and selection should be practiced was whether the yield of the progeny obtained by crossing the selected backcrossed plants with the nonrecurring parent equaled or exceeded the yield of the $F_1$ generation plants obtained by crossing the original inbred lines. Richey and Sprague (*40*) found three or four generations of back-pollinating to be sufficient for the selection of some lines whose behavior in such crosses would be equal or superior to the recurring parent. Moreover, they stated that experience also indicated that only two or three generations of selfing after back-pollinating would be required to fix the recovered lines sufficiently for a second cycle of convergent improvement.

With the immediately foregoing methods and results in mind, it is easier to follow Richey and Sprague's (*40*) discussion as to the bearing of their data on the theories for the explanation of heterosis. In 1931, when they wrote their paper, East's theory had not been advanced. They examined their data to determine whether heterosis could most logically be accounted for by the physiological-stimulation hypothesis or by the hypothesis based on at least partial dominance of genes favorable to growth. However, their remarks concerning the stimulation hypothesis could just as well apply to East's theory of divergence in function of members of allels, since according to both theories heterosis is dependent upon heterozygosity. Richey and Sprague's (*40*) data pertaining to the problem under consideration were divided into two separate categories.

In the first category were given the yields of the $F_2$ generations, the yields of the $F_1$ corn crosses back-pollinated to one parent for different numbers of generations and then crossed with the nonrecurring parent, and the yields of the $F_1$ generations obtained by crossing the original inbred lines. Selection to retain genes from the nonrecurring parent was practiced during the back-pollinating program. Richey and Sprague (*40*) compared the obtained yields with the theoretical yields calculated on the assumption that the effects of the genes are arithmetically cumulative. That this assumption probably was correct will be shown later. They pointed out that for the first backcross generation selection could not be practiced, but that for the remaining backcross generations selection for genes from the nonrecurring parent would make these selected lines more like the nonrecurring parent. Hence, according to the theory of divergence in function between members of allels, the yields of the crosses between these selected lines and the nonrecurring parent should be lower than the theoretical yields. As a matter of fact, Richey and Sprague (*40*) found the reverse to be true, as would be expected on the basis of the partial-dominance theory for the explanation of heterosis.

In the second category were discussed the yields of $F_1$ generations resulting from crossing selected inbred lines and the yields of the $F_1$ generations resulting from crossing the original inbred lines. In any one cross the two selected inbred lines resulted from a program of backcrossing to different parents. According to the theory of divergence in function between members of allels, the $F_1$ generations resulting from crossing selected lines should produce lower yields than the $F_1$ generations resulting from crossing the original inbred lines. Richey and Sprague (*40*) obtained quite the reverse result. The $F_1$ crosses $(3 \times 2^4) \times (2 \times 3^4)$ and $(3 \times 2^5) \times (2 \times 3^5)$ yielded more than the actual parental $F_1$ cross $2 \times 3$. Again, the results are those expected on the basis of the partial-dominance theory for the explanation of heterosis. However, the results do not exclude the possibility of divergence in function of members of allels accounting for some of the heterosis exhibited by these $F_1$ generations; but the results do furnish rather conclusive evidence that such a phenomenon cannot be the sole cause of heterosis. Considerable research needs to be conducted with a variety of plant material and characters before the relative importance of these two phenomena in bringing about heterosis can be evaluated.

It will be remembered that Jones (*23*), in his hypothesis explaining why the corn breeders had failed to obtain inbred lines as vigorous as the $F_1$ generations, assumed linkage between genes favorable to growth and those not so favorable. Anderson (*1*), working with a cross between *Nicotiana alata* and *N. langsdorffii*, has published data that have a bearing upon this problem. He discussed the hindrances to free recombination of characters under the headings gametic elimination, zygotic elimination, "spurious pleiotropy," and linkage. In his material the two primary causes of gametic elimination were nonviability of pollen and genes for self-sterility. Anderson's (*1, p. 682*) comments concerning zygotic elimination in species crosses may be summed up by his statement:

> The complexion of any particular $F_2$ will be determined by the particular environment in which it was grown as well as by the possibilities inherent in the germ cells which begot it. Even under the so-called standard conditions of a scientific experiment there are numerous uncontrolled variables to which any two species will react differentially. These will include such factors as time of year, sunshine, humidity, temperature, crowding, fumigation, and others. For every differentially selective factor in the environment there will tend to be a selection of $F_2$ segregates. An ideal $F_2$ is as impossible as an ideal environment.

"Spurious pleiotropy" may be of considerable importance in all breeding work involving hybridization. For example, it is rather evident that any characters entirely dependent upon the same physiological genetic processes cannot be recombined. In regard to linkage, Anderson concludes that if the number of factors affecting a character equals or exceeds one per cross-over segment, then all such characters will be tightly linked, without regard to the further restrictions imposed by frequencies. From theoretical considerations he makes the following statement (*1, p. 689*):

> If we suppose that the species differ on the average by only four genes per crossover segment (which seems a ridiculously low value) then the total hindrance is in the neighborhood of 1/2048. This means that even though we were to grow an $F_2$ so large that it occupied all the arable land on the earth, we would still be obtaining less than 1/2000 of the gene combinations possible with no linkage. In our actual $F_2$ therefore we have been obtaining only a fraction of this fraction,

and in addition there have been the further restrictions imposed by pleiotropy and gametic and zygotic elimination.

Anderson (*1*) points out that his data are in agreement with the theoretical consideration. These studies further emphasize the difficulties one may expect to encounter in attempting to obtain inbred lines as vigorous as those $F_1$ generations exhibiting considerable heterosis.

### NATURE OF THE INTERACTIONS OF GENES AS DETERMINED BY END PRODUCTS

The literature on the nature of the interactions of the genes as determined by end products is rather extensive, as in the broadest sense it includes both the physiological genetic and morphogenetic studies as well as other studies on quantitative inheritance. Also, much information can be obtained from papers on the applied phase of genetics, namely, plant and animal breeding. No attempt will be made to review all the literature having a bearing on this subject. Only a brief review covering some of the more important principles will be given. For a more extensive survey the reader is referred to Goldschmidt (*16*).

The theory of gene action as developed by Goldschmidt (*16*) has a direct bearing upon an interpretation of the data concerning the nature of the interactions of the genes differentiating quantitative characters, these interactions being determined by the measurement of end products. Goldschmidt pictures the development of the individual as due to reaction velocities in tune and postulates further that the reactions involved are catalyzed by genes. That the genes control rates of reactions (*15*, *48*) is basic to this theory. Other biological conceptions basic to Goldschmidt's theory of gene action are thresholds, timing, interrelationships of the different reactions catalyzed by the genes, and the numerical systems involved. A discussion of each of these phenomena has been given in previous publications (*16*, *37*) and need not be repeated here. However, since the present study has to do with the numerical systems involved, it may be well to review briefly some of the investigations pertaining to this phase of the matter.

At least two numerical systems are of interest. (1) There may be no interactions between the genes affecting the quantitative character. Then, the effects of these genes will be arithmetically cumulative. This is well illustrated by the work of Mangelsdorf and Fraps (*34*), who found that in corn the vitamin A units per gram increased approximately 2.25 for each additional *Y* gene.

(2) The nature of the interactions of the genes affecting the quantitative character may be such that the effects of the genes are geometrically cumulative. The methods of attacking this problem have been quite diverse. From statistical considerations, Galton (*14*) expressed the belief that the forces acting on vital phenomena tend to have constant rate effects (effects in geometric progression). East (*9*) and Groth (*17*), from genetical and statistical considerations, came to the conclusion that in certain cases the effects of genes differentiating quantitative characters are geometrically cumulative. Lindstrom (*31*), working with crosses between species of *Lycopersicon*, clearly realized that this may be true. Houghtaling (*20*), from morphogenetic studies on the size of the tomato fruit, reached the same conclusion. Sinnott (*46*), MacArthur and Butler (*33*), and Charles and Smith (*3*), from a study of the means and frequency

distributions of the parents, and of the $F_1$ and $F_2$ generations, interpreted the results from size-inheritance studies as showing that the effects of the genes differentiating the quantitative characters are geometrically cumulative. Powers (*36*, *37*), by the use of marker genes, found that the nature of the interactions between the genes carried in different regions of the chromosome and the nature of the interactions between the factors represented by generations and those carried in the different regions of the chromosome were such that the effects of these genes were geometrically cumulative. The investigations of Houghtaling (*20*) and of MacArthur and Butler (*33*) show that size of fruit in the tomato is dependent upon at least three stages of development: Locule pattern formation, increase in cell number, and cell expansion. Since these stages of development occur in the sequence given, one might expect the effects of the genes to be geometrically cumulative. Powers (*37*) found that such was the case. He termed this an interstagic type of interaction of the genes. His work showed also that the intrastagic type of interaction of the genes pertaining to number of locules was such that the effects of the genes were geometrically cumulative.

## EXPERIMENTAL CONSIDERATIONS

The experimental considerations in designing an experiment are dependent to a great extent on the objectives of the investigations. The objective of the present study was to obtain more information concerning the genetic phenomena of heterosis and dominance and concerning the nature of the interactions of the genes differentiating the quantitative characters.

Yield being a very important economic consideration in tomato production, the following characters affecting yield were selected for study: Weight per fruit, number of locules per fruit, number of fruits per centimeter of branch, height of plant, and spread of plant. It is apparent that weight of fruit and number of locules have economic importance other than through the influence they have upon yield. In obtaining weight of fruit data, 5 fruits per plant were weighed and averaged in 1938 and 10 fruits per plant in 1939. The weights were expressed in grams. As for number of locules the counts were based on 2 fruits per plant in 1938 and on 5 fruits per plant in 1939. To determine number of fruits per centimeter of branch, one representative main branch of each plant was measured for length and the fruits on that branch were counted; then the number of fruits was divided by the length of the branch expressed in centimeters. In measuring height of plant and spread of plant, care was taken to obtain measurements representative of the plant and to avoid extremes. Height is the distance from the ground level at the base of the plant to the top, and spread of the plant is the diameter measured through the main stem as the center. Both of these measurements are expressed in centimeters.

From genetic considerations and since the objective of the investigations was to obtain more information concerning the phenomena of heterosis and dominance and the nature of the interactions of the genes differentiating these characters, it is apparent that the parents, the $F_1$ generation, the $F_2$ generation, and the backcrosses to the respective parents should be included in the tests. All of this material was grown in a randomized-block experiment, there being a total of 20

blocks in each of the years 1938 and 1939. In 1938, two plots per block of each segregating generation were grown to one plot per block of the parents and $F_1$ generation. In 1939 only one plot of each was grown per block. The parents and generations were randomized within blocks, the randomization being the same for greenhouse propagation and field planting within any specified year. All operations such as transplanting were performed on the basis of blocks. Hence, since randomization was within blocks, any differences due to operations would not be confounded with the differences, such as those between generations, between parents, and between parents and generations, which the experiment was designed to study.

The parental material comprised the varieties Bonny Best, Danmark, and Johannisfeuer of *Lycopersicon esculentum* Mill. and the Red Currant variety of *L. pimpinellifolium* (Jusl.) Mill. The crosses Johannisfeuer × Danmark and Danmark × Red Currant were grown in 1938, and the crosses Johannisfeuer × Red Currant, Johannisfeuer × Bonny Best, and Johannisfeuer × Danmark were grown in 1939, Johannisfeuer × Danmark being grown during both years of the study. The purpose of the experimental design was to place the greatest emphasis on variety of plant material without entirely neglecting environmental influences as represented by years. The immediately foregoing discussion may be summarized as follows: Data for 5 quantitative characters were taken from somewhat more than 12,000 plants. The 12,000 plants represented 4 different crosses and were grown in a randomized block experiment composed of 20 blocks.

The methods employed in reducing the data were essentially the same as those given by Powers (*36*, *37*) with the exception of some minor changes, desirable because of the nature of the experiment. Only these changes need be mentioned here. Since the interest in this study lies primarily in comparisons between averages of parents and generations, the plot instead of the plant was used as the unit for reducing the data. Then, for each parent and generation the standard errors were estimated for within crosses and for within generations and parents on the basis of the averages of plots, these averages being derived from the individual plant data for the respective plots. In all tables the standard errors of the means are given. Differences having $P$ values less than 0.05 were considered as statistically significant. The symbol $B_1$, which appears both in the tables and in the text, signifies that the progeny indicated resulted from backcrossing the $F_1$ to the designated parent.

The details of the experimental design are given in another paper (*38*) dealing with stages of development and need not be repeated here.

## HETEROSIS AND DOMINANCE

### THEORETICAL CONSIDERATIONS

When interpreting and evaluating the data, it is of decided benefit to have in mind the theoretically expected behavior of the generation means. First, the $F_1$ mean will be compared with the parental means to obtain information as to whether heterosis, complete dominance, partial dominance, or no dominance exists. With this information available, it is possible to predict where the means of the backcrossed generations and the mean of the $F_2$ generation should fall in respect to the mean of the $F_1$ and the two parental means. If the $F_1$ generation shows heterosis, at least one of the backcross generations should

show heterosis also. This should be the backcross to the parent more closely approaching the $F_1$. The other backcross may or may not show heterosis, depending upon the disparity between the means of the parents. The mean of the $F_2$ should fall between the means of the backcross generations and would not be a very close approximation of the mean of the $F_1$. On the other hand, if there were no heterosis but instead some type of dominance or no dominance, theoretically the following behavior would be expected: The mean of either backcross generation should fall between the mean of the $F_1$ and the mean of the parent to which the backcross was made; and the mean of the $F_2$ should fall between the mean of the backcross generations, and it should approach more closely the mean of the $F_1$ as the degree of dominance became less. Hence, with no dominance, the $F_2$ mean should be a very close approximation of the $F_1$ mean. With these theoretical predictions in mind we are in a better position to interpret and evaluate the data.

## EXPERIMENTAL DATA

The experimental results will be discussed under five headings: (1) Weight of fruits, (2) number of locules per fruit, (3) number of fruits per centimeter of branch, (4) plant height, and (5) plant spread.

### Weight of Fruits

The means for weights of the individual fruits expressed in grams are given in table 1. For the cross Danmark × Johannisfeuer, in both 1938 and 1939, the mean of the $F_1$ generation falls between the means of the two parents but is closer to the mean of the smaller fruited parent, Johannisfeuer. The difference between the two differences involved in this latter comparison is statistically significant for 1938 but is not statistically significant for 1939. However, since these differences for both years are in the same direction, one can safely conclude that small size of fruit is partially dominant. The remaining data for this cross for 1938, aside from the fact that the means of the backcross generations and the mean of the $F_2$ fall between the means of the two parents, are not sufficiently consistent to allow any conclusion to be drawn either for or against the supposition that small size of fruit is partially dominant. The data for the cross Danmark × Johannisfeuer collected in 1939 and the data for the other crosses may be discussed together. In every case the mean of the $F_1$ lies closer to the mean of the smaller fruited parent.

For the crosses Danmark × Red Currant and Johannisfeuer × Red Currant the differences involved in the comparisons are statistically significant, whereas the differences involved in the comparisons for the crosses Danmark × Johannisfeuer and Johannisfeuer × Bonny Best do not reach statistical significance. In these crosses small size of fruit seems to be partially dominant. If such is the case, the following should be true: The mean of the backcross to the larger fruited parent should fall between the mean of the $F_1$ and the mean of the larger fruited parent; the mean of the backcross to the smaller fruited parent should fall between the mean of the $F_1$ and the mean of the smaller fruited parent; and the mean of the $F_2$ should fall between the means of the two backcross generations. The experimental data for Danmark × Johannisfeuer in 1939 and for the other three crosses are in general agreement with the theoretical expectation. Hence,

from these data it may be concluded that small size of fruit is partially dominant for the tomato crosses involved.

TABLE 1.—*Means for weight of individual fruits*

| Hybrid, generation, or parent | Mean weight of fruits in— | | Hybrid, generation, or parent | Mean weight of fruits in 1939 |
|---|---|---|---|---|
| | 1938 | 1939 | | |
| Danmark × Johannisfeuer: | *Grams* | *Grams* | Johannisfeuer × Bonny Best: | *Grams* |
| Johannisfeuer | 40.130±2.773 | 55.004±2.330 | Johannisfeuer | 55.004±2.330 |
| $B_1$ to Johannisfeuer | 45.975±1.918 | 64.040±2.260 | $B_1$ to Johannisfeuer | 63.826±2.816 |
| $F_1$ generation | 41.325±1.996 | 70.043±3.035 | $F_1$ generation | 82.019±3.119 |
| $F_2$ generation | 49.548±1.908 | 69.329±1.934 | $F_2$ generation | 81.336±3.847 |
| $B_1$ to Danmark | 43.970±1.810 | 78.079±2.405 | $B_1$ to Bonny Best | 90.736±3.011 |
| Danmark | 54.015±2.675 | 87.934±2.017 | Bonny Best | 119.616±3.037 |
| Danmark × Red Currant: | | | Johannisfeuer × Red Currant: | |
| Red Currant | .915± .032 | | Red Currant | .903± .029 |
| $B_1$ to Red Currant | 2.095± .068 | | $B_1$ to Red Currant | 2.265± .086 |
| $F_1$ generation | 5.480± .318 | | $F_1$ generation | 6.682± .134 |
| $F_2$ generation | 5.380± .196 | | $F_2$ generation | 6.066± .318 |
| $B_1$ to Danmark | 16.105± .552 | | $B_1$ to Johannisfeuer | 18.948± .805 |
| Danmark | 51.170±3.548 | | Johannisfeuer | 55.004±2.330 |

## NUMBER OF LOCULES PER FRUIT

The means for number of locules per fruit are listed in table 2. The data for the Danmark × Johannisfeuer and the Johannisfeuer × Bonny Best crosses are very similar and hence may be discussed together. In no case is the $F_1$ mean significantly different from the mean of the fewer loculed parent. It would seem that small number of locules is completely dominant. This conclusion is substantiated by the means of the backcross to the fewer loculed parent. In fact, for 1939 the mean of the backcross to the fewer loculed parent in both of these crosses is smaller than the mean of the fewer loculed parent. The means of the backcrosses to the many-loculed parent in all cases fall between the mean of the $F_1$ and the mean of the many-loculed parent. The means of the $F_2$ in all cases fall between the means of the backcrosses, and approximate but are somewhat greater than the means of the $F_1$. The consistency of the data both for years and for crosses leads the writer to put considerable confidence in the conclusion that small number of locules is completely dominant in the crosses Danmark × Johannisfeuer and Johannisfeuer × Bonny Best.

TABLE 2.—*Means for number of locules per fruit*

| Hybrid, generation, or parent | Mean locules per fruit in— | | Hybrid, generation, or parent | Mean locules per fruit in 1939 |
|---|---|---|---|---|
| | 1938 | 1939 | | |
| Danmark × Johannisfeuer: | *Number* | *Number* | Johannisfeuer × Bonny Best: | *Number* |
| Danmark | 5.475±0.057 | 6.183±0.065 | Bonny Best | 6.318±0.069 |
| $B_1$ to Danmark | 5.575± .064 | 5.898± .097 | $B_1$ to Bonny Best | 5.452± .077 |
| $F_1$ generation | 5.500± .086 | 6.051± .096 | $F_1$ generation | 6.329± .111 |
| $F_2$ generation | 6.595± .118 | 6.826± .150 | $F_2$ generation | 6.781± .208 |
| $B_1$ to Johannisfeuer | 7.500± .100 | 7.344± .136 | $B_1$ to Johannisfeuer | 7.034± .162 |
| Johannisfeuer | 9.125± .091 | 9.028± .084 | Johannisfeuer | 9.028± .084 |
| Danmark × Red Currant: | | | Johannisfeuer × Red Currant: | |
| Red Currant | 2.050± .014 | | Red Currant | 2.034± .004 |
| $B_1$ to Red Currant | 2.205± .015 | | $B_1$ to Red Currant | 2.183± .016 |
| $F_1$ generation | 2.395± .018 | | $F_1$ generation | 2.517± .029 |
| $F_2$ generation | 2.570± .022 | | $F_2$ generation | 2.886± .078 |
| $B_1$ to Danmark | 3.473± .037 | | $B_1$ to Johannisfeuer | 4.356± .140 |
| Danmark | 5.405± .068 | | Johannisfeuer | 9.028± .084 |

Likewise, the data for the crosses Danmark × Red Currant and Johannisfeuer × Red Currant are very similar and therefore may be discussed together. The $F_1$ means lie between the means of the respective parents, but in both cases materially closer to the mean of the fewer loculed parent. It would appear that small number of locules is partially dominant. The means of the backcrosses to the parents having smaller number of locules lie between the $F_1$ means and the means of the parents having smaller number of locules. The means of the other backcrosses lie between the $F_1$ means and the means of the parents having a greater number of locules. The means of the $F_2$ fall between the means of the backcross generations and approach the $F_1$ means rather closely but are somewhat larger. The data for Danmark × Red Currant and Johannisfeuer × Red Currant are in agreement with the conclusion that small number of locules is partially dominant. Again the consistency of the data allows one to have considerable confidence in the validity of this deduction.

### Number of Fruits per Centimeter of Branch

The means for number of fruits per centimeter of branch are given in table 3. First, consider the cross Danmark × Johannisfeuer. The mean of the $F_1$ for 1938 falls between the means of the parents, indicating partial dominance of fewer fruits per centimeter of branch. The means of the backcrosses and the mean of the $F_2$ support this conclusion. For this same cross in 1939 the mean of the $F_1$ and the mean of the backcross to Johannisfeuer are not significantly different from the mean of Johannisfeuer, the parent having smaller number of fruits. This would indicate complete dominance of small number of fruits per centimeter of branch. The mean of the $F_2$ and the mean of the backcross to the parent having a greater number of fruits per centimeter of branch are in agreement with this conclusion.

For the cross Johannisfeuer × Bonny Best the difference between the means of the parents is not very great. The fact that the means of all the hybrid generations, including the $F_1$, fall between the means of the two parents is rather conclusive evidence that dominance is partial.

Table 3.—*Means for number of fruits per centimeter of branch*

| Hybrid, generation, or parent | Mean fruits per cm. branch in— | | Hybrid, generation, or parent | Mean fruits per cm. branch in 1939 |
|---|---|---|---|---|
| | 1938 | 1939 | | |
| Danmark × Johannisfeuer: | *Number* | *Number* | Johannisfeuer × Bonny Best: | |
| Johannisfeuer | 0.216±0.006 | 0.220±0.009 | | *Number* |
| $B_1$ to Johannisfeuer | .225± .004 | .201± .008 | Bonny Best | 0.187±0.008 |
| $F_1$ generation | .243± .007 | .214± .007 | $B_1$ to Bonny Best | .197± .010 |
| $F_2$ generation | .261± .006 | .239± .010 | $F_1$ generation | .192± .007 |
| $B_1$ to Danmark | .320± .005 | .264± .012 | $F_2$ generation | .191± .008 |
| Danmark | .410± .012 | .337± .014 | $B_1$ to Johannisfeuer | .197± .007 |
| Danmark × Red Currant: | | | Johannisfeuer | .220± .009 |
| Danmark | .417± .012 | | Johannisfeuer × Red Currant: | |
| $B_1$ to Danmark | .404± .010 | | Johannisfeuer | .220± .009 |
| $F_1$ generation | .359± .009 | | $B_1$ to Johannisfeuer | .241± .009 |
| $F_2$ generation | .428± .011 | | $F_1$ generation | .302± .015 |
| $B_1$ to Red Currant | .423± .011 | | $F_2$ generation | .303± .016 |
| Red Currant | .441± .017 | | $B_1$ to Red Currant | .393± .024 |
| | | | Red Currant | .548± .030 |

Considering the $F_1$ for the cross Danmark × Red Currant, the following facts regarding number of fruits per centimeter of branch become apparent. The mean of the $F_1$ is smaller than the mean of either parent; the mean of the backcross to Danmark falls between the mean of the $F_1$ and the mean of Danmark; and the mean of the $F_2$ and the mean of the backcross to Red Currant fall between the mean of the $F_1$ and the mean of the Red Currant parent. These facts are typical of heterosis. That this is an expression of heterosis is apparent from a consideration of the length of internode as determined by the distance between leaf axils in the $F_1$ as compared with the same character in the parents. The distance between leaf axils was considerably greater in the $F_1$ than was the distance between leaf axils in either parent. This fact is reflected in the spread and height data, which will be given later. The greater length of the internodes of the $F_1$ would result in a greater number of centimeters per cluster and would be expected to result in fewer fruits per centimeter of branch length. Accordingly, the heterosis noted is probably due to the vegetative vigor of the plants.

For the cross Johannisfeuer × Red Currant, the $F_1$ mean falls between the means of the two parents; the mean of the backcross to the Johannisfeuer parent falls between the means of the $F_1$ and of the Johannisfeuer parent; and the mean of the backcross to the Red Currant parent and the $F_2$ mean fall between the means of the $F_1$ and of the Red Currant parent. Hence, fewer fruits per centimeter of branch length is partially dominant.

These data clearly demonstrate that heterosis and dominance, insofar as inheritance of the character number of fruits per centimeter of branch is concerned, are dependent upon both the cross and the environmental conditions as represented by years.

### Plant Height

The means for plant height, expressed in centimeters, are listed in table 4. The crosses Danmark × Johannisfeuer, Danmark × Red Currant, and Johannisfeuer × Red Currant may be discussed together. In these cases the means of the $F_1$ exceed the means of all other hybrid generations and the means of the parents. Moreover, in all of these crosses the means of the backcross generations fall between their respective $F_1$ means and those of the parents to which they were backcrossed. In all these crosses the means of the $F_2$ generations fall between the means of their respective backcross generations. One other fact concerning the data should be noted. With the exception of the $B_1$ to Danmark mean in 1939 (for the cross Danmark × Johannisfeuer) all of the hybrid generation means exceed the means of their respective parents. From these data it may be concluded that all of these crosses show heterosis for plant height.

For the cross Johannisfeuer × Bonny Best, the $F_1$ mean falls between the means of the two parents, but slightly closer to the mean of the taller parent. The difference involving two differences ($F_1$—Johannisfeuer)—(Bonny Best—$F_1$), in this comparison is not statistically significant. For the cross Johannisfeuer × Bonny Best, plant height approaches intermediacy or no dominance. The magnitudes of the means for the other hybrid generations are in accord with these conclusions.

TABLE 4.—*Means for plant height*

| Hybrid, generation, or parent | Mean plant height in— | | Hybrid, generation, or parent | Mean plant height in 1939 |
|---|---|---|---|---|
| | 1938 | 1939 | | |
| Danmark × Johannisfeuer: | *Centimeters* | *Centimeters* | Johannisfeuer × Bonny Best: | *Centimeters* |
| Danmark | 35.40±0.617 | 35.57±0.427 | Johannisfeuer | 39.43±0.495 |
| $B_1$ to Danmark | 39.38± .687 | 38.45± .490 | $B_1$ to Johannisfeuer | 41.66± .675 |
| $F_1$ generation | 43.80± .942 | 41.21± .677 | $F_1$ generation | 43.67± .606 |
| $F_2$ generation | 41.38± .622 | 39.56± .740 | $F_2$ generation | 43.61± .868 |
| $B_1$ to Johannisfeuer | 41.80± .683 | 41.07± .516 | $B_1$ to Bonny Best | 45.28± .566 |
| Johannisfeuer | 38.95± .851 | 39.43± .495 | Bonny Best | 46.87± .556 |
| Danmark × Red Currant: | | | Johannisfeuer × Red Currant: | |
| Red Currant | 27.90± .676 | | Red Currant | 36.79± .665 |
| $B_1$ to Red Currant | 35.73± .666 | | $B_1$ to Red Currant | 39.77± .762 |
| $F_1$ generation | 44.45±1.202 | | $F_1$ generation | 43.49± .832 |
| $F_2$ generation | 35.93± .646 | | $F_2$ generation | 39.93± .855 |
| $B_1$ to Danmark | 38.88± .600 | | $B_1$ to Johannisfeuer | 40.59± .524 |
| Danmark | 34.85± .704 | | Johannisfeuer | 39.43± .495 |

### PLANT SPREAD

The means for plant spread, expressed in centimeters, are given in table 5. The data for all of the crosses may be considered together. Without exception the means of the $F_1$ generations exceeded the means of all other hybrid generations and the means of the parents in their respective crosses. Also, in every cross the mean of the backcross to the parent with the greatest spread exceeded the mean of the parent with the greatest spread. The means of the backcrosses to the parents with the smaller spread, in all cases, fell between the means of the respective $F_1$ generations and those of the parents with the smaller spread. Therefore, we may conclude that all crosses show heterosis for plant spread, heterosis being particularly pronounced in crosses including Red Currant.

TABLE 5.—*Means for plant spread*

| Hybrid, generation, or parent | Mean plant spread in— | | Hybrid, generation, or parent | Mean plant spread in 1939 |
|---|---|---|---|---|
| | 1938 | 1939 | | |
| Danmark × Johannisfeuer: | *Centimeters* | *Centimeters* | Johannisfeuer × Bonny Best: | *Centimeters* |
| Danmark | 66.65±1.916 | 107.45±2.651 | Bonny Best | 128.05±1.605 |
| $B_1$ to Danmark | 77.05±2.243 | 130.46±1.938 | $B_1$ to Bonny Best | 139.52±2.151 |
| $F_1$ generation | 94.70±4.379 | 155.32±1.625 | $F_1$ generation | 151.06±1.933 |
| $F_2$ generation | 84.25±2.483 | 139.65±2.626 | $F_2$ generation | 146.47±1.970 |
| $B_1$ to Johannisfeuer | 88.15±2.664 | 153.49±1.996 | $B_1$ to Johannisfeuer | 147.73±2.056 |
| Johannisfeuer | 79.20±3.509 | 144.59±2.103 | Johannisfeuer | 144.59±2.103 |
| Danmark × Red Currant: | | | Johannisfeuer × Red Currant: | |
| Danmark | 65.85±2.263 | | Johannisfeuer | 144.59±2.103 |
| $B_1$ to Danmark | 93.68±2.294 | | $B_1$ to Johannisfeuer | 173.93±2.597 |
| $F_1$ generation | 137.55±4.943 | | $F_1$ generation | 198.38±2.202 |
| $F_2$ generation | 105.98±2.720 | | $F_2$ generation | 173.97±2.113 |
| $B_1$ to Red Currant | 120.68±3.531 | | $B_1$ to Red Currant | 179.46±1.980 |
| Red Currant | 106.65±4.475 | | Red Currant | 159.98±1.302 |

## NATURE OF INTERACTIONS OF GENES

### THEORETICAL CONSIDERATIONS

The data concerning the nature of the interactions of the genes have a bearing upon two of the possible numerical systems involved. It is important from the standpoint of both genetics and plant breeding to

know whether the effects of the genes are arithmetically or geometrically cumulative. The formulas used in obtaining the theoretical means are the same as those described by Powers and Lyon (*38*) and need not be repeated here. As previously pointed out (*38*), the method employing these formulas is not always discriminative. This was found to be true in the case of many of the data collected in this study. In fact, the method was found to be discriminative for only part of the data on weight of individual fruits and for only part of those on number of locules.

## EXPERIMENTAL DATA

### Weight of Fruits

The obtained and theoretical means for weight of individual fruits are given in table 6. Very little if any information can be obtained concerning the nature of the interactions of the genes for the crosses Danmark × Johannisfeuer and Johannisfeuer × Bonny Best, because the two sets of theoretical means are very nearly equal. However, such is not the case with the crosses Danmark × Red Currant and Johannisfeuer × Red Currant. There are only very small differences

TABLE 6.—*Obtained and theoretical means for weight of individual fruits*

| Hybrid, year, and generation | Mean weight of fruits | | |
|---|---|---|---|
| | Obtained | Theoretical | |
| | | Geometric | Arithmetic |
| Danmark × Johannisfeuer: | | | |
| *1938* | *Grams* | *Grams* | *Grams* |
| $B_1$ to Johannisfeuer | 45.975±1.918 | 40.723 | 40.728±1.717 |
| $F_2$ generation | 49.548±1.908 | 43.863 | 44.199±1.411 |
| $B_1$ to Danmark | 43.970±1.810 | 47.246 | 47.670±1.634 |
| *1939* | | | |
| $B_1$ to Johannisfeuer | 64.040±2.260 | 62.070 | 62.524±1.881 |
| $F_2$ generation | 69.329±1.934 | 69.794 | 70.756±1.730 |
| $B_1$ Danmark | 78.079±2.405 | 78.480 | 78.989±1.924 |
| Johannisfeuer × Bonny Best: | | | |
| *1939* | | | |
| $B_1$ to Johannisfeuer | 63.826±2.816 | 67.167 | 68.512±1.912 |
| $F_2$ generation | 81.336±3.847 | 81.565 | 84.665±1.849 |
| $B_1$ to Bonny Best | 90.736±3.011 | 99.049 | 100.818±2.264 |
| Danmark × Red Currant: | | | |
| *1938* | | | |
| $B_1$ to Red Currant | 2.095± .068 | 2.239 | 3.198± .076 |
| $F_2$ generation | 5.380± .196 | 6.124 | 15.761± .214 |
| $B_1$ to Danmark | 16.105± .552 | 16.745 | 28.325± .757 |
| Johannisfeuer × Red Currant: | | | |
| *1939* | | | |
| $B_1$ to Red Currant | 2.265± .086 | 2.456 | 3.793± .046 |
| $F_2$ generation | 6.066± .318 | 6.862 | 17.318± .114 |
| $B_1$ to Johannisfeuer | 18.948± .805 | 19.171 | 30.848± .449 |

between the obtained means and the theoretical means calculated on the assumption that the effects of the genes differentiating weight of fruit are geometrically cumulative. It is rather apparent that the fit is very poor between the obtained means and those calculated on the assumption that the effects of the genes differentiating weight of fruit are arithmetically cumulative. Even though the fit is fairly good between the obtained means and the theoretical means based on

the geometrically cumulative hypothesis, it should be noted that in both crosses these theoretical means are consistently larger than the obtained means. It is quite evident that these differences cannot be attributed to the errors of random sampling. One cannot draw the conclusion from these data that the effects of the genes differentiating weight of fruit are strictly geometrically cumulative. Some unknown quantity is exerting an influence.

### Number of Locules per Fruit

The obtained and theoretical means for number of locules are given in table 7. Again for the crosses Danmark × Johannisfeuer and Johannisfeuer × Bonny Best, a comparison between obtained and theoretical means is of little value in determining the nature of the interactions of the genes as determined by measurements of end products. Again the difficulty lies in the nondiscriminative nature of the method employed. However, such is not the case with the crosses Danmark × Red Currant and Johannisfeuer × Red Currant. The

Table 7.—*Obtained and theoretical means for number of locules*

| Hybrid, year, and generation | Mean locules per fruit | | |
|---|---|---|---|
| | Obtained | Theoretical | |
| | | Geometric | Arithmetic |
| Danmark × Johannisfeuer: | | | |
| *1938* | *Number* | *Number* | *Number* |
| $B_1$ to Danmark | 5.575±0.064 | 5.487 | 5.488±0.052 |
| $F_2$ generation | 6.595± .118 | 6.235 | 6.400± .054 |
| $B_1$ to Johannisfeuer | 7.500± .100 | 7.084 | 7.313± .066 |
| *1939* | | | |
| $B_1$ to Danmark | 5.898± .097 | 6.117 | 6.117± .058 |
| $F_2$ generation | 6.826± .150 | 6.724 | 6.828± .058 |
| $B_1$ to Johannisfeuer | 7.344± .136 | 7.391 | 7.540± .068 |
| Johannisfeuer × Bonny Best: | | | |
| *1939* | | | |
| $B_1$ to Bonny Best | 5.452± .077 | 6.323 | 6.324± .065 |
| $F_2$ generation | 6.781± .208 | 6.914 | 7.001± .066 |
| $B_1$ to Johannisfeuer | 7.034± .162 | 7.559 | 7.679± .075 |
| Danmark × Red Currant: | | | |
| *1938* | | | |
| $B_1$ to Red Currant | 2.205± .015 | 2.216 | 2.223± .011 |
| $F_2$ generation | 2.570± .022 | 2.824 | 3.061± .015 |
| $B_1$ to Danmark | 3.473± .037 | 3.598 | 3.900± .026 |
| Johannisfeuer × Red Currant: | | | |
| *1939* | | | |
| $B_1$ to Red Currant | 2.183± .016 | 2.263 | 2.276± .013 |
| $F_2$ generation | 2.886± .078 | 3.284 | 4.024± .020 |
| $B_1$ to Johannisfeuer | 4.356± .140 | 4.767 | 5.773± .035 |

fit between the obtained means and those calculated on the assumption that the effects of the genes are arithmetically cumulative is not good. The fit between the obtained means and the means calculated on the assumption that the effects of the genes are geometrically cumulative is considerably closer. But again the calculated means for both crosses are considerably larger than the obtained means; and again it is apparent that some unknown quantity or quantities are influencing

the results. From these data we cannot safely conclude that the effects of the genes are strictly geometrically cumulative. By the use of marker genes Powers (*36, 37*) has shown that in the *Lycopersicon* material studied the results were those expected on the theory that the effects of the interactions of the genes influencing both weight of fruit and number of locules are geometrically cumulative.

## DISCUSSION AND CONCLUSIONS

### HETEROSIS AND DOMINANCE

#### Plant-Breeding Methods Utilizing Heterosis and Dominance

Before taking up the relation that the foregoing data have to a tomato-breeding program, it may be well to consider some of the plant-breeding methods that have been developed to utilize heterosis and dominance. These methods include the following: (1) Inbreeding, followed by hybridization; (2) production of $F_1$ hybrids, three-way hybrids, double hybrids, advanced-generation hybrids, and inbred line $\times$ variety hybrids; (3) production of synthetic varieties; (4) predicting yields of hybrids from inbred lines; (5) improvement of inbred lines.

Shull (*42, 43, 44, 45*), East (*6, 7, 8*), Jones (*23, 24*), and East and Hayes (*11*) did the early work leading to the modern methods of breeding crops in which the utilization of heterosis is a primary consideration. They demonstrated the advantages of inbreeding followed by hybridization. Hence, the production of inbred lines and the determination of the combining ability (ability to produce high-yielding hybrids) of these inbred lines became a standard procedure for corn breeders.

Problems concerning the most efficient means of utilizing these inbred lines soon arose. Shull (*43*) was the first to suggest growing the $F_1$ generation between two inbred lines of corn to produce the commercial crop. However, the poor yield and quality of seed produced by inbred lines of corn are obstacles to the use of $F_1$ hybrids. To overcome these difficulties Jones (*24*) proposed the use of double crosses. Soon after this, three-way crosses came into use. Jones' idea was to cross two $F_1$ generations of different inbred parentage and to use seed thus produced for planting the commercial crop. This method has proved very practical and is now in general use throughout the United States. In 1930 Kiesselbach (*26*) published results showing that yields from advanced-generation hybrids were equal to those from the original double cross. As the name implies, this method involves crosses between generations subsequent to the $F_1$. For example, the two $F_2$ generations obtained from the $F_1$ generations making up the original double cross would be hybridized to produce an advanced-generation double cross. The use of inbred line $\times$ variety hybrids for the production of the commercial crop was first suggested by Lindstrom (*30*). This method involves crossing an inbred line and a variety, and in some cases satisfactory yields have been obtained.

All of the foregoing methods developed to utilize heterosis in corn production have a common disadvantage, namely, that the farmer cannot save seed for the production of next year's crop from the current crop. To overcome this difficulty Hayes and Garber (*19*) suggested using inbred lines to produce synthetic varieties. So far no

outstanding synthetic varieties have been produced. It seems that synthetic varieties offer promise, and hence the production of such varieties merits more consideration than it has received in the past. This fact is emphasized by the findings of Wright (*49*), Neal (*35*), and Kiesselbach (*26*). Wright (*49*) has shown that "a random-bred stock derived from *n* inbred families will have 1/*n*th less superiority over its inbred ancestry than the first cross or a random-bred stock from which the inbred families might have been derived without selection." Neal (*35*) found that for 10 single hybrids, 4 three-way hybrids, and 10 double hybrids the agreement was very good between the obtained and theoretical means (calculated by the use of Wright's formula). Kiesselbach (*27*), in 1933, reported the results of tests made over a period of years to determine the comparative amounts of yield reduction in the second generation of various kinds of crosses, including single, double, 8-line, and 16-line hybrids. He reached the following conclusions:

> The decrease in yield of any hybrid in the $F_2$ generation is equal to half the difference in yield between its $F_1$ and the mean yield of the open pollinated parents. This will also apply to any advanced generation in the absence of selection. This reduced yield is due to a reduction in the number of favorable growth factors as a consequence of close breeding. In any orthodox hybrid in which the two parents are regularly built up from equal numbers of lines this reduction tends to be inversely proportional to the number of lines involved.

Thus Wright (*49*), working with animals, and Kiesselbach (*27*), working with plants, drew similar conclusions. From these investigations it seems rather safe to conclude that the amount of decrease in yield of grain in *Zea mays* accompanying inbreeding may be predicted by Wright's (*49*) formula, and therefore it should be possible to produce a high-yielding synthetic variety, provided a sufficient number of inbred lines having high combining ability are available.

One of the biggest problems of the corn breeder is ascertaining the combining ability of inbred lines when used in single, three-way, and double crosses. The obvious method is to make the possible crosses between the inbred lines and then test these crosses for yielding ability. Where large numbers of inbred lines are available, this method is almost prohibitive owing to the large amount of labor, time, and land required for testing. Therefore, methods materially reducing these requirements are needed. Jenkins and Brunson (*22*) pointed out that, for producing double crosses, multiple crosses, or synthetic varieties, the most valuable lines are those that cross well with a large number of inbred lines, i. e., those that produce, on the average, the best hybrids when tested with a rather wide range of germ plasm. Jenkins and Brunson (*22*) also reported the results of studies to determine whether this wide range of germ plasm might not be supplied by crossing with a mixed collection of varieties, or possibly by crossing with an individual variety. From these studies they concluded that crosses with open-pollinated varieties may be used efficiently in the preliminary testing of new lines. Following up this work, Jenkins (*21*) found that the information obtained from comparisons of inbred-line variety crosses may be utilized to advantage in estimating the performance of double crosses among these lines. Jenkins (*21*) also gave a method of estimating the yields of double crosses, based on genetic considerations. He pointed out that—

> In any double cross the genes of each of the four parental lines are united only with allelomorphs of the two lines which entered the double cross from the opposite

parent. In the double cross $WX \times YZ$, therefore, it would seem that the crosses *WY*, *WZ*, *XY*, and *XZ* better represent the hybrid combinations actually occurring than do all six of the possible combinations among these lines.

Following this suggestion, Doxtator and Johnson (*5*) demonstrated that highly significant differences in yielding ability can be found in double crosses resulting from the use of single-cross parents produced from four inbred lines, and that by the appropriate use of single-cross data the highest yielding double-cross combination may be predicted. Hence, it would seem that the methods developed by Jenkins (*21*) would greatly expedite testing the combining ability of inbred lines.

From the foregoing discussions, it is apparent that the improvement of inbred lines, particularly as to yielding ability and quality of seed, would aid materially in solving many of the corn-breeding problems. Convergent improvement was developed by Richey (*39*) as a method of increasing the yielding ability of inbred lines and at the same time maintaining their combining ability with certain other inbred lines. The procedure for this method was as follows: Two inbred lines that combined exceptionally well were crossed to produce an $F_1$ generation; then the $F_1$ generation was backcrossed to each inbred line; selection for vigor and characters of the nonrecurring parent was practiced in these two series of backcrosses; these selections of any one series were again backcrossed to the inbred line occurring in that series, and again, from the progeny, individuals possessing vigor and other desirable characters of the nonrecurring parent were selected. This procedure with the two series of backcrosses was continued until the yields of the crosses between the selected lines and the nonrecurring parent equaled or exceeded the yields of the $F_1$ generation obtained from crossing the original inbred lines. Richey and Sprague (*40*) found that three or four backcrossings were sufficient to produce hybrids whose yields were approximately equal to the yields of the $F_1$ crosses between the foundation inbred lines. Moreover, in these backcrossed generations there was indicated a permanent improvement of 13 to 15 percent over the recurring parent after allowing for differences in the degree of inbreeding. Then, as Richey and Sprague (*40*) pointed out, inasmuch as this excess presumably is due to the retention of additional dominant genes from the nonrecurring parent, as a result of selection among the progeny resulting from back-pollinating, the recovered lines are heterozygous for these genes and hence must be inbred to put at least some of these genes in the homozygous condition. After this has been accomplished the breeder is in a position to start a second cycle of convergent improvement. In summarizing the results from their tests with corn, Richey and Sprague (*40*) drew the following conclusions: In addition to the larger yields of the recovered lines, improvement has been achieved in ability to resist lodging and in the amount of pollen shed; yellow endosperm has been substituted for white and clear pericarp for red, all without changing significantly the behavior of the lines in crosses; convergent improvement, suggested originally from theoretical considerations as a means of improving selfed lines of corn without interfering with their behavior in hybrid combination, so far has been found successful; the results suggest that this method also may provide a means by which the yields of $F_1$ crosses between selfed lines can be raised to an even higher level. From the results obtained by Richey and Sprague, it seems safe to

conclude that convergent improvement is a practical method of improving yielding ability and other desirable characters of inbred lines.

### Application of Heterosis and Dominance to Tomato Breeding

In a tomato-breeding program it is apparent that the standard methods of breeding self-fertilized crops can and should be used. However, it is not so apparent that the methods developed for the improvement of cross-fertilized crops can be used advantageously in a tomato-breeding program. Information concerning heterosis and dominance of the more important economic characters of tomatoes is essential to the solution of this problem. The data reported in this paper give some of this information.

Small size of fruit, as determined by weight, was found to be partially dominant. Nearly the same situation exists in regard to number of locules, small number of locules being either completely or partially dominant. Partial dominance or heterosis was exhibited for smaller number of fruits per centimeter of branch. From these results it would seem that the tomato breeder cannot utilize heterosis to increase size of fruit, number of locules, or number of fruits per centimeter of branch. However, if smaller fruits or fruits with a smaller number of locules are desired, these can be obtained in hybrids by crossing with varieties or strains possessing these characters. Heterosis for increased height and spread of plant was obtained. Also, it will be remembered (*38*) that some of the crosses discussed in this paper exhibited decided heterosis for earliness of maturity. This was particularly true for those crosses having Red Currant as one of the parents. In regard to the characters studied, it may be concluded that the greatest benefits due to heterosis will come from increased plant size and increased earliness. Some of the increase in plant size should result in increased plant yields. It seems that increases in plant size and earliness are sufficient to warrant an intensive study of the possibility of utilizing heterosis in the commercial production of tomatoes.

These considerations indicate that the tomato breeder should employ those fundamental principles established by geneticists and breeders who have already attempted to take advantage of heterosis. For example, in building up inbred lines and varieties to be used in crosses, foundation stock as genetically diverse as possible should be used. The decided heterosis for increased plant size and for increased earliness exhibited by the crosses between *Lycopersicon esculentum* and *L. pimpinellifolium* amply demonstrates the advantages of such a method. During the development of these inbred lines and varieties the breeder and geneticist should not forget the necessity of obtaining information concerning the inheritance of the economically important characters. The breeding program can be pursued with much less expense and with much more certainty of success if information concerning the inheritance of these characters is available. The truth of this assertion is verified by the fact that all of the important advances in methods of corn breeding have been based on fundamental genetic principles. Also, the value of genetic information is well illustrated by the studies concerning weight of fruit, number of locules per fruit, and number of fruits per centimeter of branch length reported in this article. Since small weight of fruit, small

number of locules, and small number of fruits per centimeter of branch are at least partially dominant, it is evident that the requirements desired in the commercial product should be striven for in the inbred lines, especially if *L. pimpinellifolium* has been used as foundation stock. On the other hand, such characters as earliness and size of plant can be obtained by taking advantage of heterosis, provided the inbred lines or varieties used in the crosses possess good combining ability. It is very apparent that further advances in methods of breeding and the utilization of the methods we now have will be dependent upon genetic information concerning the crop and knowledge of fundamental genetic principles.

In building up strains of tomatoes to be used in crosses, the backcross method and Richey's (*39*) convergent-improvement method will undoubtedly be of value. In using these methods caution should be exercised in order that material too similar genetically does not result. In certain material, to avoid this danger inherent in the backcross method, selection within $F_2$ and later generations could be practiced to advantage.

## Relation of Heterosis and Dominance

In this paper it has been assumed that from the standpoint of physiological genetics there is no fundamental difference between the phenomena of heterosis and dominance. Some of the data reported in a previous article (*38*) pertain to this assumption. These data are presented in tables 8 and 9.

Table 8.—*Means of number of days from seeding to first complete change of color of any fruit for the cross Danmark × Johannisfeuer*

| Generation or parent | Mean period from seeding to first complete change of color in— | |
|---|---|---|
| | 1938 | 1939 |
| | *Days* | *Days* |
| Johannisfeuer | 164.9±0.900 | 136.1±0.838 |
| $B_1$ to Johannisfeuer | 165.0± .463 | 133.3± .787 |
| $F_1$ generation | 165.6± .617 | 132.4± .440 |
| $F_2$ generation | 166.4± .437 | 134.9± .727 |
| $B_1$ to Danmark | 167.6± .382 | 134.8± .983 |
| Danmark | 170.0± .489 | 137.7± .632 |

Table 9.—*Means of number of days from first fruit set to first complete change of color of any fruit for 1939*

| Hybrid, generation, or parent | Mean period from first fruit set to first complete change of color | Hybrid, generation, or parent | Mean period from first fruit set to first complete change of color |
|---|---|---|---|
| Johannisfeuer × Red Currant: | *Days* | Danmark × Johannisfeuer: | *Days* |
| Johannisfeuer | 45.2±0.483 | Johannisfeuer | 45.2±0.483 |
| $B_1$ to Johannisfeuer | 44.0± .498 | $B_1$ to Johannisfeuer | 47.6± .564 |
| $F_1$ generation | 43.5± .472 | $F_1$ generation | 48.0± .516 |
| $F_2$ generation | 44.5± .608 | $F_2$ generation | 49.9± .412 |
| $B_1$ to Red Currant | 47.1± .535 | $B_1$ to Danmark | 50.3± .621 |
| Red Currant | 49.3± .501 | Danmark | 54.6± .563 |

From table 8 it can be seen that there are no significant differences between the 1938 means of number of days from seeding to first complete change of color in the case of Johannisfeuer, the $B_1$ to Johannisfeuer, and the $F_1$ generation. This shows that for 1938 smaller number of days from seeding to first complete change of color of any fruit is completely dominant. In 1939 the means of the $F_1$ generation and of the $B_1$ to Johannisfeuer were significantly smaller than the means of Johannisfeuer and of Danmark. Hence, smaller number of days from seeding to first complete change of color showed heterosis in 1939. Since the genotypes were constant for the two years, it is evident that the environment as represented by years determined whether the character should exhibit heterosis or dominance.

The means of number of days from first fruit set to first complete change of color of any fruit are listed in table 9. From this table it can be seen that for Johannisfeuer $\times$ Red Currant the means of the $F_1$, the $B_1$ to Johannisfeuer, and the $F_2$ generations are smaller than the mean of either parent. In these comparisons, the differences involving the mean of the $F_1$ are statistically significant. From these facts it may be concluded that the character smaller number of days from first fruit set to first complete change of color of any fruit exhibits heterosis. For Danmark $\times$ Johannisfeuer the means of all the generations are between the means of the two parents. Clearly smaller number of days from first fruit set to first complete change of color of any fruit is partially dominant. Since in this case the environmental influence has been controlled (randomized-block experiment with 20 replications) we may conclude that whether the character small number of days from first fruit set to first complete change of color of any fruit exhibits heterosis or dominance is determined by the genotype.

Hence, as the foregoing data indicate, in one case the environment and in another case the genotype determined whether a character should exhibit heterosis or partial dominance. This evidence is rather conclusive in support of the supposition that heterosis and dominance represent degrees of identical phenomena and are dependent upon the same physiological genetic processes.

## NATURE OF INTERACTIONS OF GENES

Although genetics has contributed much to the advancement of plant breeding, there is need for considerably more work on the inheritance of the quantitative characters. This is particularly true for studies dealing with the nature of the interaction of the genes affecting the quantitative characters.

The corn breeders have accumulated considerable data having a bearing upon the nature of the interaction of the genes as determined by the measurement of an end product, namely, yield. Their data indicate that the effects of the genes differentiating yield in corn are arithmetically cumulative. In fact those formulas used in predicting theoretical decreases in yield accompanying inbreeding (*27*, *35*, *40*, *49*) are based on the assumption that the effects of the genes differentiating yield are arithmetically cumulative. The fact that such good fits between the obtained and theoretical means were secured by these formulas would indicate that this assumption is correct. However, before such a conclusion is drawn tests should be made to determine

whether the formulas are discriminative. In other words, would the obtained and theoretical data fit just as well if the theoretical calculations had been made on the assumption that the effects of the genes differentiating the quantitative characters are geometrically cumulative? To answer this question the theoretical $F_2$ means were calculated from Neal's (*35*) data for corn by using the formulas given in table 10.

These theoretical means, together with the obtained means, are listed in table 11.

TABLE 10.—*Formulas for calculating theoretical $F_2$ mean yields of Zea mays from Neal's (35) data*

| Hybrids | Arithmetic mean | Geometric mean |
|---|---|---|
| Single | $\frac{2\overline{F}_1+\overline{P}_1+\overline{P}_2}{4}$ | Antilogarithm of $\left(\frac{2\log\overline{F}_1+\log\overline{P}_1+\log\overline{P}_2}{4}\right)$ |
| 3-way | $\frac{6\overline{F}_1+\overline{P}_1+\overline{P}_2+\overline{P}_3}{9}$ | Antilogarithm of $\left(\frac{6\log\overline{F}_1+\log\overline{P}_1+\log\overline{P}_2+\log\overline{P}_3}{9}\right)$ |
| Double | $\frac{12\overline{F}_1+\overline{P}_1+\overline{P}_2+\overline{P}_3+\overline{P}_4}{16}$ | Antilogarithm of $\left(\frac{12\log\overline{F}_1+\log\overline{P}_1+\log\overline{P}_2+\log\overline{P}_3+\log\overline{P}_4}{16}\right)$ |

TABLE 11.—*Obtained and theoretical $F_2$ mean yields of Zea mays, the calculations being based on Neal's (35) data*

$F_2$ MEAN YIELDS OF ZEA MAYS

| Cross | $F_2$ mean yield | | | Cross | $F_2$ mean yield | | | Cross | $F_2$ mean yield | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Obtained | Arithmetic [1] | Geometric [2] | | Obtained | Arithmetic [1] | Geometric [2] | | Obtained | Arithmetic [1] | Geometric [2] |
| Single hybrid: | *Bu.* | *Bu.* | *Bu.* | 3-way hybrid: | *Bu.* | *Bu.* | *Bu.* | Double hybrid (con.): | *Bu.* | *Bu.* | *Bu.* |
| 3×$R_3$ | 45.7 | 40.4 | 35.6 | ($R_3$×3)×25 | 49.0 | 51.7 | 46.2 | ($R_3$×6)×(3×25) | 55.1 | 55.6 | 51.4 |
| $R_3$×6 | 47.0 | 43.0 | 38.5 | (3×25)×$R_3$ | 44.7 | 48.1 | 43.7 | (23×$R_3$)×($M_{12}$×6) | 52.6 | 54.4 | 50.5 |
| $R_3$×25 | 43.7 | 41.5 | 36.3 | ($R_3$×25)×6 | 56.9 | 54.4 | 48.9 | (3×6)×(23×25) | 57.4 | 55.2 | 51.9 |
| $R_3$×26 | 34.8 | 38.5 | 34.4 | (6×25)×$R_3$ | 46.5 | 48.8 | 44.9 | ($R_3$×25)×($M_{12}$×23) | 45.2 | 52.9 | 49.0 |
| 3×25 | 41.7 | 47.7 | 41.7 | Double hybrid: | | | | (3×$R_3$)×(6×23) | 56.9 | 55.6 | 51.6 |
| 3×26 | 44.7 | 46.3 | 40.8 | ($R_3$×6)×(23×25) | 52.0 | 54.7 | 50.9 | | | | |
| 6×26 | 52.6 | 47.9 | 43.4 | ($M_{12}$×$R_3$)×(6×25) | 50.0 | 49.6 | 46.5 | | | | |
| $M_{12}$×$R_3$ | 40.9 | 39.7 | 34.9 | (3×$R_3$)×(23×25) | 58.7 | 57.1 | 52.3 | | | | |
| $R_3$×23 | 44.5 | 41.6 | 37.0 | (6×25)×(3×$R_3$) | 58.2 | 56.9 | 52.4 | | | | |
| 23×26 | 46.5 | 46.5 | 41.7 | (23×$R_3$)×(3×25) | 53.6 | 51.4 | 47.9 | | | | |

ANALYSIS SUMMARY OF SINGLE HYBRIDS FOR OBTAINED AND ARITHMETIC MEANS

| Source of variation | Degrees of freedom | Sum of squares | Variance | *F* value | *P* value |
|---|---|---|---|---|---|
| Crosses | 9 | 239.908000 | 26.656444 | 3.837914 | <0.05 |
| Means | 1 | 4.050000 | 4.050000 | .583107 | >.05 |
| Crosses × means | 9 | 62.510000 | 6.945556 | | |
| Total | 19 | 306.468000 | | | |

[1] The *P* values for determining the fit between the obtained and arithmetic means were greater than 0.05.
[2] The *P* values for determining the fit between the obtained and geometric means were less than 0.01.

Some explanation concerning the method of treatment of these data should be given. To determine whether the obtained means were in

agreement with either group of the theoretical means, the analysis of variance was applied to each group of hybrids. The method used is shown at the bottom of table 11. The data included in this illustration are the means obtained for the single hybrids and the means calculated on the assumption that the effects of the genes differentiating yields are arithmetically cumulative. In using this method one should examine the detailed data to ascertain whether a few extreme deviates occur. If this is done there should be no misinterpretation of the data.

From table 11 it can be seen that the agreement is generally good between the obtained data and the theoretical data calculated on the assumption that the effects of the genes differentiating yield in these inbred lines of corn are arithmetically cumulative. On the other hand, the disparity between the obtained data and those calculated on the assumption that the effects of the genes are geometrically cumulative is in general too great to be accounted for by the probable errors of random sampling. These findings are supported by the data of Richey and Sprague (*40*) and by those of Kiesselbach (*27*). It should be pointed out that in applying the above formulas to data for three-way and double hybrids, if the possible $F_1$ hybrids differ materially in yield the data from the highest yielding combination (*5*, *21*) should be used; otherwise erroneous conclusions may be drawn. Where the possible $F_1$ hybrids do not differ greatly in yield this consideration seems to be merely academic.

It will be remembered that in the present study with tomatoes the agreement between the obtained data and those calculated on the assumption that the effects of the genes are geometrically cumulative was closer than was the agreement between the obtained data and those calculated on the assumption that the effects of the genes are additive. However, in neither instance was the agreement so close that the differences noted could be attributed to the errors of random sampling. The characters involved were weight of fruits and number of locules per fruit. By using the method involving marker genes, Powers (*36*, *37*) has shown that the results obtained in a study of these two characters are those expected if the effects of the genes differentiating these two characters and associated with the marker genes are geometrically cumulative. Hence, it seems rather evident, as previously pointed out by Powers (*37*), from theoretical considerations and a study of other data, that more than one type of interaction of the genes differentiating quantitative characters must exist when measurements of end products are involved.

## SUMMARY

The studies here reported included as parental material the Bonny Best, Danmark, and Johannisfeuer varieties of *Lycopersicon esculentum*, and the Red Currant variety of *L. pimpinellifolium*. The breeding was carried to the second hybrid and two backcross generations and in addition the study included the $F_1$ generation and both parents.

Small size of tomato fruit as determined by weight was found to be partially dominant.

Small number of locules was found to be either completely or partially dominant.

Partial dominance or heterosis was exhibited for smaller number of fruits per unit length of branch.

Heterosis obtained for increased height and spread of plant.

It was concluded that the increase in plant size and the previously determined earliness, due to heterosis (*38*), are sufficient to warrant an intensive study of the possibility of utilizing this phenomenon in the commercial production of tomatoes.

The agreement between the obtained means and those calculated on the assumption that the effects of the genes are geometrically cumulative was closer than was the agreement between the obtained means and those calculated on the assumption that the effects of the genes are arithmetically cumulative. However, in neither instance was the agreement so close that the differences noted could be attributed to the errors of random sampling.

Evidence has been presented showing that from the standpoint of physiological genetics heterosis and dominance are merely different degrees of the same phenomena.

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# INHERITANCE OF STAGES OF EARLINESS IN AN INTERSPECIFIC CROSS BETWEEN LYCOPERSICON ESCULENTUM AND L. PIMPINELLIFOLIUM[1]

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## INTRODUCTION

In 1936, at the Cheyenne Horticultural Field Station, Cheyenne, Wyo., an interspecific cross was made between the tomato varieties Johannisfeuer (*Lycopersicon esculentum* Mill.) and Red Currant (*L. pimpinellifolium* (Jusl.) Mill.) in an attempt to combine certain important characters of Johannisfeuer with the desirable earliness of Red Currant. The $F_1$, $F_2$, and backcross generations, together with the parents, were grown in 1937, and the results on studies of the inheritance of earliness are reported by Powers and Lyon.[3] The $F_3$ generation and advanced backcross generation, together with the $F_1$ generation, $F_2$ generation, and parents, were turned over to the author in 1938 for an investigation of the inheritance of earliness.

## METHODS

This study included the parental lines, the $F_1$ generation, the $F_2$ generation, $F_3$ families arising from self-fertilized plants resulting from backcrossing the $F_1$ generation to Johannisfeuer, and $F_3$ families arising from self-fertilized $F_2$ plants. The backcross plants and the $F_2$ plants giving rise to the last two classes had been selected for earliness of maturity in 1937. The seed was planted in flats in the greenhouse on April 21, 1938. After germination the seedlings were spotted off in flats in strain rows, which were randomized according to Tippett's randomization tables[4] to eliminate possible positional effects in the greenhouse. The strains and generations were transplanted into the field June 1, 1938, in rows, 12 plants to the row, each row being replicated 4 times and randomized as to position within a block. Ten blocks of 48 rows each were used, each block containing both parents, the $F_1$ and $F_2$ generations, 4 strains of the selfed generations of the backcross, and 4 strains of selected $F_3$'s. This made possible the testing of 40 strains of the selfed generation of the backcross and 40 strains of $F_3$'s in a randomized block design,[5] using a total population of 5,760 plants.

On July 13, 1938, when the large-scale breeding population was entirely in bloom, a severe hailstorm occurred, inflicting considerable

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damage on the entire population. The effect of the hail was uniform over the entire field planting, and only stubs of plant stems were left above ground. Since hailstorms inflicting such damage occur frequently during this season in many portions of the central Western States, they must be considered as part of the environment in any breeding program. Under conditions that are present in such an environment, inheritable earliness is confused with ability to recover after hail damage.

For convenience and ease of analysis, earliness in tomatoes may be divided into various stages. Powers and Lyon [6] have recognized the following stages and have found they behave as genetic characters and can be treated as such in studies of quantitative inheritance: (1) Period from planting to first bloom; (2) period from first bloom to first fruit set; (3) period from first fruit set to first complete color change. In the experiment herein reported the occurrence of hail delimited additional stages, so that five different stages are employed in this study. The following is a list of the stages, together with the criteria that delimit them in actual field practice:

(1) *Period from planting to first bloom before hail.*—The day that the corolla of the first flower opened was recorded for each plant, and the number of days from the planting date was computed for this stage of earliness.

(2) *Period from planting to first bloom after hail.*—The same procedure was followed as in stage 1, as all plants were pruned back to a uniform basis and all flower buds were removed immediately after damage occurred.

(3) *Period from first bloom after hail to first fruit set.*—When a definite swelling of an ovary in a flower occurred, the date was recorded and the number of days from first bloom was computed for this stage of earliness.

(4) *Period from first fruit set to first complete color change.*—The date of first complete color change was recorded for each plant when the green coloring on a fruit had disappeared, and the number of days from first fruit set to first complete color change was computed for this stage of earliness.

(5) *Period from planting to first complete color change.*—This stage of earliness represents a summation of stages 2, 3, and 4.

The mean values of rows were used to establish differences between strains or generations. The data thus obtained were then subjected to analyses of variance, and the interaction between strains and replications was used as an estimate of error. Odds as great as or greater than 19 : 1 against the deviations noted being due to the errors of random sampling were accepted as being statistically significant.

## RESULTS

Table 1 gives the *F* values for testing the statistical significance of differences between strain means and of differences between replication means. The differences between the strain means will be discussed under their respective stage headings.

### PERIOD FROM PLANTING TO FIRST BLOOM BEFORE HAIL DAMAGE

From table 1, it may be concluded that, for period 1, in no case could the difference between strain means be accounted for by chance. In all cases the *F* values were of sufficient magnitude to give probability values as small as or less than 0.01.

[6] See footnote 3.

TABLE 1.—*F values for testing the statistical significance of differences between strain means and of differences between replication means*

BETWEEN STRAINS [1]

| Block No. | *F* value for indicated period | | | | |
|---|---|---|---|---|---|
| | (1) Planting to first bloom (before hail) | (2) Planting to first bloom (after hail) | (3) First bloom after hail to first fruit set | (4) First fruit set to first color change | (5) Planting to first color change |
| 1 | 10.92 | 4.23 | 2.74 | 1.73 | 3.20 |
| 2 | 14.45 | 4.29 | 1.16 | 4.85 | 5.10 |
| 3 | 18.75 | 6.08 | 1.67 | 8.86 | 13.84 |
| 4 | 7.45 | 5.83 | .37 | 5.21 | 9.60 |
| 5 | 5.99 | 5.58 | 1.61 | 3.21 | 4.92 |
| 6 | 5.54 | 3.57 | 1.89 | 10.46 | 11.02 |
| 7 | 14.33 | 6.50 | 2.53 | 9.37 | 11.73 |
| 8 | 3.70 | 3.07 | 2.21 | 2.78 | 3.65 |
| 9 | 20.85 | 8.66 | 1.12 | 5.14 | 8.98 |
| 10 | 5.10 | 5.74 | 1.39 | 6.38 | 10.23 |

BETWEEN REPLICATIONS [2]

| | | | | | |
|---|---|---|---|---|---|
| 1 | 0.01 | 0.26 | 2.05 | 3.20 | 3.49 |
| 2 | 1.77 | 2.23 | .76 | .72 | 1.11 |
| 3 | .39 | .81 | .94 | 4.68 | 5.48 |
| 4 | 6.62 | 1.25 | 1.00 | 5.01 | 6.46 |
| 5 | .52 | 2.26 | 1.18 | 1.19 | 1.07 |
| 6 | 2.62 | 2.36 | .80 | 2.18 | 3.79 |
| 7 | .99 | 3.91 | .01 | 5.45 | 8.78 |
| 8 | .79 | .24 | .90 | 2.87 | 1.64 |
| 9 | .13 | 1.87 | 2.20 | .51 | .75 |
| 10 | .80 | 1.38 | 1.60 | .40 | .52 |

[1] 2.84 is the 1-percent level and 2.09 is the 5-percent level for values of *F* with 11 degrees of freedom for strains and 33 degrees of freedom for the interaction of strains and replications, which was used as error.
[2] 4.44 is the 1-percent level and 2.89 is the 5-percent level for values of *F* with 3 degrees freedom for replications and 33 degrees of freedom for the interaction of strains and replications, which was used as error.

Before hail, the Red Currant parent exhibited a mean number of days to first bloom of $74.55 \pm 0.444$ days from the planting date. Johannisfeuer, the later parent, bloomed in $81.54 \pm 0.271$ days, making it possible to demonstrate a parental mean difference of $6.99 \pm 0.520$ days. The $F_1$ generation bloomed in $76.13 \pm 0.446$ days and differed from the arithmetic mean of the two parents ($78.04 \pm 0.260$ days) by $1.91 \pm 0.516$ days. The mean difference is over three times its standard error and therefore significant. The simple geometric mean (77.97 days) obtained from the means of the parents does not conform to the observed mean of the $F_1$ generation as a mean difference of 1.84 days is demonstrable. Also, from the foregoing data it is apparent that the $F_1$ generation differs significantly from the earlier parent, as the difference is $1.58 \pm 0.629$ days. In other words, although the geometric mean obtained from the means of the two parents more closely approaches the observed mean of the $F_1$ generation than does the arithmetic mean obtained from the means of the two parents, dominance is a factor and must be considered. This agrees with data of Powers [7] for number of locules and fruit size.

The observed mean of the $F_2$ generation for date of first bloom before hail was $75.76 \pm 0.408$ days. By using Wright's formula as quoted by Powers,[7] which takes into consideration dominance in geometric in-

[7] POWERS, LEROY. STUDIES ON THE NATURE OF THE INTERACTIONS OF THE GENES DIFFERENTIATING QUANTITATIVE CHARACTERS IN A CROSS BETWEEN LYCOPERSICON ESCULENTUM AND L. PIMPINELLIFOLIUM. Jour. Genet. 39: [139]-170. 1939.

heritance as shown by the $F_1$ generation, one may obtain a predicted $F_2$ mean of $77.04 \pm 0.261$ days. While the standard error of the predicted $F_2$ mean is in itself subject to error and represents only the best approximation available, the difference between predicted and observed $F_2$ means of $1.28 \pm 0.484$ is statistically significant. Therefore, in considering the $F_2$ generation, the viewpoint given above of a geometrically inherited character taking into account dominance is not substantiated.

The data show that it is possible by selection within the $F_2$ generation to obtain $F_3$ strains that are at least 8 days earlier than the Red Currant parent in regard to date of first bloom. Table 2 lists 10 such $F_3$ strains, giving strain means as well as the standard errors.

TABLE 2.—*Tabulation of the mean number of days from planting to first bloom before hail of parents, $F_1$ and $F_2$ generations, and early selected $F_3$ strains*

| Parent or strain No. | Mean | Error | Generation or strain No. | Mean | Error |
|---|---|---|---|---|---|
| Parent: | *Days* | *Days* | Generation: | *Days* | *Days* |
| Red Currant | 74.55 | ±0.444 | $F_1$ | 76.13 | ±0.446 |
| Johannisfeuer | 81.54 | ±.271 | $F_2$ | 75.76 | ±.408 |
| Selected $F_3$ strain: | | | Selected $F_3$ strain: [1] | | |
| 26–10 | 66.02 | ±3.830 | 98–12 | 62.50 | ±2.423 |
| 40–7 | 57.25 | ±1.691 | 144–3 | 61.31 | ±2.188 |
| 60–15 | 56.29 | ±3.529 | 210–8 | 60.87 | ±1.119 |
| 72–23 | 66.13 | ±1.425 | 211–4 | 64.19 | ±3.309 |
| 97–16 | 63.04 | ±3.589 | 216–1 | 66.44 | ±1.192 |

[1] These strains were at least 8 days earlier than the Red Currant parent in the period from planting to first bloom before hail.

## PERIOD FROM PLANTING TO FIRST BLOOM AFTER HAIL DAMAGE

From table 1 it is clearly demonstrable that any differences shown between strain means are valid and significant. In all of the 10 blocks, one is able to demonstrate significance above the 1-percent point for the differences between means of the strains, or, in all cases the odds are greater than 99:1 against the occurrence by chance of such a distribution of strain means.

After the hailstorm the Red Currant parent bloomed in $98.26 \pm 0.195$ days from the date of planting. The later parent, Johannisfeuer, bloomed in $102.02 \pm 0.189$ days, and it is possible after hail damage to demonstrate a mean difference between parental lines of only $3.76 \pm 0.272$ days in blooming dates. When one recalls the difference of $6.99 \pm 0.520$ days that was demonstrable before hail occured, one may conclude that the expression of this genetic difference was decreased, in this experiment, by damage due to hail.

The $F_1$ generation shows a mean number of days to first bloom of $97.89 \pm 0.209$. This mean conforms neither to the arithmetic mean ($100.14 \pm 0.136$ days) obtained from the means of parental lines nor to the simple geometric mean (100.12 days) obtained in the same manner. It is impossible to demonstrate conclusively that the $F_1$ generation is significantly earlier than the Red Currant parent, since a difference of only $0.37 \pm 0.286$ day is shown.

The observed $F_2$ mean was $99.13 \pm 0.194$ days, which is very close to the predicted $99.00 \pm 0.125$ days based on the assumption that genes differentiating this stage of development are geometrically cumulative. Refined analysis is not necessary to show that the difference between these two means is not statistically significant.

The selected $F_3$ strains that had shown earliness in regard to date of first bloom before hail gave no indications whatsoever of blooming early after hail damage occurred. There was no indication of early blooming after hail in any $F_3$ strain or in any strain of the selfed generation of the backcross, and the $F_1$ generation in general showed the only indications in the population of blooming earlier than the Red Currant parent.

### PERIOD FROM FIRST BLOOM TO FIRST FRUIT SET

The Red Currant variety of tomato requires a period from first bloom to first fruit set of $4.25 \pm 0.090$ days, while the Johannisfeuer variety requires a shorter period of $3.58 \pm 0.121$ days. The parental strains thus exhibit a significant mean difference of $0.67 \pm 0.151$ day. The $F_1$ generation mean ($3.88 \pm 0.109$ days) lies between those of the parents and differs significantly from the mean of the Red Currant parent, the mean difference being $0.37 \pm 0.141$ day. The difference between the Johannisfeuer parent and the $F_1$ generation ($0.30 \pm 0.163$ day) closely approaches statistical significance. There is, however, no significant difference demonstrable between the $F_1$ generation and the arithmetic mean ($3.91 \pm 0.075$ days) calculated from the means of the parents or between the $F_1$ generation and the geometric mean (3.90 days) calculated from the means of the parents.

The $F_2$ generation, with a mean of $4.05 \pm 0.094$ days for this stage of earliness, conforms to both the arithmetic and geometric means calculated from the means of the two parents, as no significant differences are involved. It also conforms to the predicted $F_2$ value ($3.89 \pm 0.067$ days) considering dominance of the heterozygote, although in this case the $F_1$ generation mean lies between the means of the two parents.

By a comparison of any two strain means, it is not possible to demonstrate that statistically significant differences between strains exist within any particular block. (See table 1.) However, directional effects between genotypes are constant within blocks; and because of the large population involved, with 40 replications within parents and within generations, errors are reduced to a minimum. Accepting, then, the errors obtained by the analysis of variance within parents and generations, it is possible to demonstrate a small but significant difference between inbred lines. However, the differences exhibited in this stage of earliness in this particular cross in the year 1938 were so small as to be negligible in a practical consideration of earliness in general. In other years, under different environmental conditions, when the character would include another factor, blossom drop, it might prove to be highly important.

### PERIOD FROM FIRST FRUIT SET TO FIRST COMPLETE COLOR CHANGE

From table 1, it can be seen that the *F* values for testing significance of mean differences for the stage from first fruit set to first complete color change are statistically significant for 9 of the 10 blocks. A mean of $49.81 \pm 0.480$ days was obtained for the Red Currant variety of tomato, while the calculated mean for the Johannisfeuer variety was $52.09 \pm 0.397$ days. A parental mean difference of $2.28 \pm 0.623$ days is thus shown. The $F_1$ generation shows heterosis, with a generation mean of $46.07 \pm 0.520$ days, and clearly can conform in no

way to either the arithmetic mean (50.95±0.311 days) or the geometric mean (50.94 days) obtained from the means of the parents.

The $F_2$ mean of 47.76±0.430 days conforms closely to the predicted mean of 48.44±0.311 days obtained by use of Wright's formula. The difference between means of 0.68±0.531 day is not statistically significant. It is possible by selection within the $F_2$ generation to obtain $F_3$ strains that are at least 6 days earlier than the Red Currant parent in the period from first fruit set to first complete color change. Table 3 lists 10 such selected $F_3$ strains, together with their standard errors.

TABLE 3.—*Tabulation, in mean number of days, for the period from first fruit set to first complete color change of the parents, the $F_1$ and $F_2$ generations, and the early selected $F_3$ strains*

| Parent or strain No. | Mean | Error | Generation or strain No. | Mean | Error |
|---|---|---|---|---|---|
| Parent: | *Days* | *Days* | Generation: | *Days* | *Days* |
| Red Currant | 49.81 | ±0.480 | $F_1$ | 46.07 | ±0.520 |
| Johannisfeuer | 52.09 | ±.397 | $F_2$ | 47.76 | ±.430 |
| Selected $F_3$ strain: | | | Selected $F_3$ strain: [1] | | |
| 60–15 | 42.81 | ±1.758 | 134–1 | 43.17 | ±1.782 |
| 65–8 | 42.02 | ±.995 | 144–3 | 42.02 | ±.405 |
| 104–2 | 42.94 | ±1.580 | 173–10 | 42.58 | ±1.750 |
| 106–15 | 40.38 | ±1.035 | 205–2 | 42.94 | ±.821 |
| 115–1 | 40.56 | ±.410 | 211–4 | 41.42 | ±.764 |

[1] These strains were at least 6 days earlier than the Red Currant parent in the period from first fruit set to first complete color change.

## PERIOD FROM PLANTING TO FIRST COMPLETE COLOR CHANGE

As one would expect from table 1, if significance is obtained for "between strains" in any of the various stages of earliness and no compensating directional effect between stages is in evidence, significance must of necessity be present in the summation of all stages from planting to first complete color change. It will be noted in stage 5 (see table 1) that the odds are greater than 99: 1 in all blocks against the occurrence by chance of such a distribution of strain means. Table 4 represents a summation of all the data on stages of earliness for the parents, the $F_1$ generation, and the $F_2$ generation.

It is shown in all cases except stage 3, where only minute differences are demonstrable, that the differences between parents are all in the same direction. The final mean difference exhibited between the inbred lines is 5.43±0.654 days and is less than the mean difference shown in the same parents up to the date of first bloom before hail damage occurred. In years when injury does not occur,[8] greater differences between parents may be expected than are noted here.

The $F_1$ generation in stage 5 shows marked heterosis in earliness as a result of summation of effects shown in stages 2 and 4. The mean of the $F_2$ generation lies between the means of the $F_1$ generation and the earlier parent, as it did in stage 4, where heterosis in the earliness of the $F_1$ generation was also noted. The mean of the $F_2$ generation is clearly predictable, as it is in all stages of earliness after hail damage occurred, in which the calculations are based on the assumptions that the effects of the genes are geometrically cumulative and that dominance as exhibited by the $F_1$ generation must be taken into account.

The $F_3$ strains that showed earliness when the period from planting to first complete color change was used as a criterion were those

[8] See footnote 3.

strains that exhibited earliness in a particular stage. In some cases earliness in one stage was compensated by lateness in another stage, and the result was a strain of no value as far as earliness in general is concerned.

It is of interest to examine the results of the analyses of variance for "between replications." Whether position in the field materially affects these stages of earliness may be determined from the corresponding *F* values. In 11 cases the *F* values (see table 1) are of sufficient magnitude to give *P* values as low as or lower than 0.05. In the character, period from first fruit set to first complete color change, four blocks showed significant *F* values and, in all cases, this was reflected in the same blocks by significance for "between replications" in the summation of all stages from planting to first complete color change.

TABLE 4.—*Complete analysis of earliness, showing generation means in days with their standard errors for the various stages of earliness*

| Stage of earliness | Red Currant parent | Johannisfeuer parent | Mean difference between parents | $F_1$ generation | $F_2$ generation | Mean difference between actual $F_2$ means and predicted $F_2$ means according to Wright's formula |
|---|---|---|---|---|---|---|
| | *Days* | *Days* | *Days* | *Days* | *Days* | *Days* |
| (1) Planting to first bloom before hail [1] | 74. 55±0. 444 | 81. 54±0. 271 | 6. 99±0. 520 | 76. 13±0. 446 | 75. 76±0. 408 | 1. 29±0. 484 |
| (2) Planting to first bloom after hail | 98. 26± . 195 | 102. 02± . 189 | 3. 76± . 272 | 97. 89± . 209 | 99. 13± . 194 | . 13± . 231 |
| (3) First bloom to first fruit set | 4. 25± . 090 | 3. 58± . 121 | . 68± . 151 | 3. 88± . 109 | 4. 05± . 094 | . 16± . 115 |
| (4) First fruit set to first color change | 49. 81± . 480 | 52. 09± . 397 | 2. 28± . 623 | 46. 07± . 520 | 47. 76± . 430 | . 68± . 531 |
| (5) Date planted to date of first color change | 152. 17± . 509 | 157. 60± . 410 | 5. 43± . 654 | 147. 69± . 528 | 150. 81± . 473 | . 43± . 578 |

[1] Severe hail damage occurred at this particular time leaving only plant stems above ground. All subsequent stages of earliness are confused with recovery effects.

Since in 11 cases the *F* values for "between replications" were statistically significant, it is apparent that stages of earliness are affected by the environmental influences represented by position in the greenhouse or field, or both. Further, from the fact that a majority of the statistically significant *F* values of the replications fall within the period from first fruit set to first color change and the entire time from planting to first color change, one may conclude that as regards this cross the stage from first fruit set to first color change is more affected by position in the field than are the other stages of earliness.

## SUMMARY

A population of 5,760 plants of a cross between *Lycopersicon esculentum* Mill. and *L. pimpinellifolium* (Jusl.) Mill., which included both parents, the $F_1$ generation, and the advanced generations, was grown at Cheyenne, Wyo., in 1938. Earliness was broken up into its constituent stages, and the number of days for each stage was recorded for each plant.

Before hail damage the parents showed a difference of 7 days between means in the period from planting to the date of first bloom.

The mean of the $F_1$ generation was significantly higher than that of the earlier parent and differed significantly from both arithmetic and geometric means of the parents. The actual $F_2$ mean did not conform to the predicted $F_2$ mean based on the assumptions that the effects of the genes are geometrically cumulative and that dominance as exhibited by the $F_1$ generation must be taken into account. It is possible, by selection of $F_2$ plants, to obtain $F_3$ strains that are earlier than either parent in this stage of earliness.

After hail damage, there was a difference of only 4 days between the means of the parents in date of first bloom. The $F_3$ strains that showed earliness in date of first bloom before hail showed no indications of earliness in date of first bloom after hail. The period from planting of seed to date of first bloom after hail was found to be shorter for the $F_1$ generation than for either of the parents or for any other generation. The $F_2$ mean is clearly predictable in this stage of earliness if one assumes that the effects of the genes are geometrically cumulative and that dominance as exhibited by the $F_1$ generation must be taken into account.

Only minute differences are demonstrable in the period from first bloom to first fruit set after hail, and the differences are found to be so small as to be negligible in a practical breeding program.

Significant differences are demonstrable between the parents in the period from first fruit set to first complete color change. The $F_1$ generation shows heterosis in this stage of earliness, and the $F_2$ generation mean is predictable if one assumes cumulative gene effects, taking into account dominance as shown by the $F_1$ generation. It is possible by selection to obtain $F_3$ strains that are earlier than either parent in this stage of earliness.

Any differences noted in earliness, when number of days from planting of the seed to first complete color change is the criterion, are merely the result of a summation of stage differences as regards the present study.

# THE BUFFER CAPACITY OF THE BLOOD OF THE SIXTH-INSTAR SOUTHERN ARMYWORM (PRODENIA ERIDANIA)[1]

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## INTRODUCTION

It is well known that biochemical reactions take place in buffered media and that minor changes in hydrogen-ion concentration can affect profoundly the course of reactions. In the blood of man a variation of more than ±0.1 pH from the normal 7.4 is considered pathological, and a change greater than ±0.4 pH is usually fatal. The pH of the blood is maintained within these narrow limits by a system of buffers whose action is well understood. The buffer effect of the various constituents of the blood, as well as its buffer capacity under both normal and pathological conditions, has been studied by Peters and Van Slyke (*14*)[2] and others.

The blood of several marine invertebrates has also been found to be well buffered. Redfield et al. (*16*) summarized the early work in this field and showed that hemocyanin was the main buffer in the blood of *Limulus polyphemus*.

The buffer capacity of the blood of only a few insects seems to have been investigated, but the blood of those studied is well buffered. The limits within which the pH of insect bood can range without resulting in the death of the insect appear not to have been determined. The variation from species to species and between individuals of the same species, however, is rather large. The pH of the blood of several species, as determined by Kocian and Špaček (*10*), Bodine (*4*), Ludwig (*11*), Babers (*1*), and others, varied between 6.0 and 7.3. Kocian and Špaček found that in the same species the blood from individual insects might vary as much as 0.4 pH.

The blood of *Dytiscus marginalis* L. was alkaline to litmus, according to Barratt and Arnold (*2*), and remained so when an equal volume of 0.03 N hydrochloric acid was added, but it became acid when an equal volume of 0.1 N acid was used. The same authors found the blood of *Hydrophilus piceus* (L.) to be alkaline to litmus but neutral when an equal volume of 0.025 N acid was added. Craig and Clark (*7*) titrated the blood of *Pieris rapae* L. and *Heliothis armigera* (Hbn.) (*H. obsoleta* (F.))[3] with 0.1 N acid and alkali and determined the buffer capacity of the two bloods. These authors give the pH of the blood of *P. rapae* larvae as 7.17. This value is of interest because of the relative scarcity of reports of alkaline insect blood, and also because

[1] Received for publication March 5, 1941.

[2] Italic numbers in parentheses refer to Literature Cited, p. 189.

[3] *Heliothis obsoleta* was used in the reference cited, whereas the name currently in use is *H. armigera* (Hbn.)

Fink (*9*), using colorimetric methods, found the blood of this larva to vary between pH 6.2 and 6.4. Using the glass electrode, the present writer has found (unpublished results) the average pH of the blood of this insect to be 6.52. Brecher (*5*) found the blood of *P. brassicae* (L.) to be well buffered. She attributed the buffering power to phosphates, which she found to be present in 0.02-molar concentration. Duval and Portier (*8*), using a number of species, found the hydrogen-ion concentration of the blood to be very stable. They also attributed the buffering power to the presence of phosphates. The blood of *Bombyx mori* L. had an acidity equivalent to 0.46 gm. of oxalic acid per 100 cc. when Nazari (*13*) titrated samples to phenolphthalein with 0.1 N potassium hydroxide.

At the present time large numbers of the southern armyworm (*Prodenia eridania* (Cram.)) are being used in various physiological and toxicological investigations. A knowledge of the buffer capacity of the blood of the larvae of this insect and the effect of nicotine vapor on its blood pH has become desirable. The present report is the result of such a study.

## MATERIALS AND METHODS

Mature, actively feeding sixth instars that had been reared in greenhouse cages on turnip and collard foliage were used. Blood was collected by snipping a proleg while it was held under mineral oil. By this treatment loss of carbon dioxide was held at a minimum, and the darkening of the blood that occurs on exposure to air was greatly retarded. When unfixed blood was used in this manner the cells coagulated rapidly, but it was thought that more drastic changes might occur if heat-fixed blood were used. Hydrogen-ion determinations were made on pooled samples of definite volume by the use of a small, ordinary bulb-type glass electrode in connection with a saturated calomel half-cell. The pH was read directly from a calibrated potentiometer dial after a zero reading had been obtained by the use of a standard buffer; the dial could be read to about $\pm 0.02$ pH. Small increments of acid or alkali were added from a microburette, and the resulting pH was read. Titration curves were plotted in which the molar concentration of acid or alkali was the ordinate and the corresponding pH the abscissa.

The buffer capacity, $\beta$, is defined by Van Slyke (*17*) as the "differential ratio dB/dpH, expressing the relation between the increment (in gram-equivalents per liter) of strong base B added to a buffer solution and the resultant increment in pH. Increment of strong acid is equivalent to a negative increment of base, or $-$dB." Since the increment of pH obtained when acid is added is also negative, the value $\beta$ is always positive. If sufficiently small increments of alkali are used, $\beta$ can be calculated directly. However, since $\beta$ is equal to the tangent of the titration curve at the corresponding pH, it is usually more convenient to obtain it in this manner, and this method was used in the determinations. Theoretical discussions of buffers and buffer capacities are given by Van Slyke (*17*), Peters and Van Slyke (*14*), and Clark (*6*).

The carbon dioxide content was determined both on pooled samples and on the blood of individual larvae by the manometric method of Van Slyke and Neill (*18*) as described by Peters and Van Slyke (*15*).

## RESULTS

The pH of the blood as drawn under oil varied between 6.50 and 6.75, with an average of 6.65. A composite titration curve from the several experiments is shown in figure 1. The buffer-capacity curve in figure 2 was obtained by plotting the several values of $\beta$

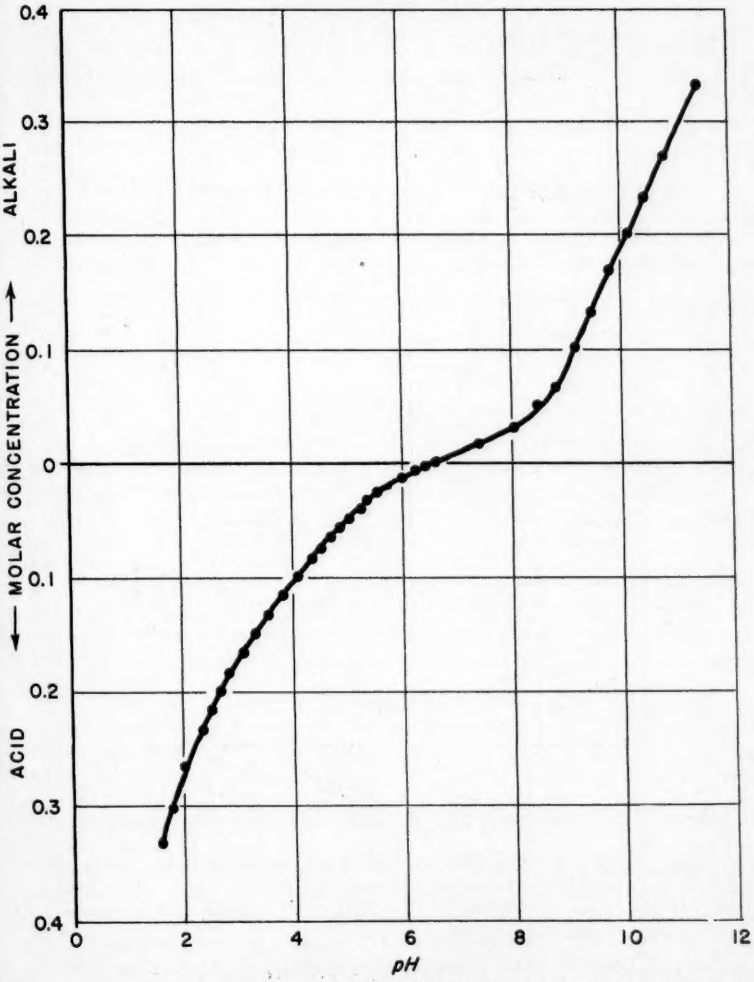


FIGURE 1.—Titration curve for the hemolymph of sixth-instar southern armyworms.

obtained from the titration curve against the corresponding pH value. The total carbon dioxide content of the blood as drawn varied between 8.40 and 13.45 volumes percent, with a mean of 10.03, for 10 analyses, or about 4.51 millimoles per liter.

Bishop (*3*) has found the dissociation constant, pK′, for the blood of honeybee larva to be 6.1, the same value as that given by Peters and Van Slyke for human serum. If this value holds for the blood of the southern armyworm, the ratio of bicarbonate to carbonate as obtained from the Henderson-Hasselbalch (*14*) equation

$$\text{pH}=\text{pK}'+\log\frac{[\text{BHCO}_3]}{[\text{H}_2\text{CO}_3]} \tag{1}$$

ranges from 3.55 to 1.

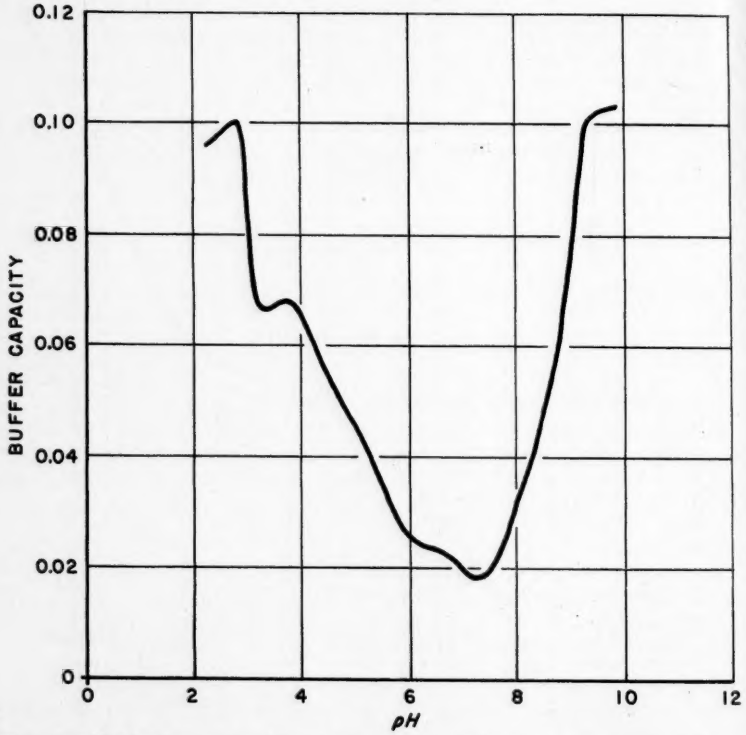


FIGURE 2.—Buffer capacity of the hemolymph of sixth-instar southern armyworms

The solubility coefficient, $\alpha$, of carbon dioxide in human serum at 38° C. is 0.92 times that of water. If the same ratio holds for larval blood, $\alpha$ at 25° will be 0.695. From the Henderson-Hasselbalch equation in the forms

$$mM[\text{H}_2\text{CO}_3]=0.0591\alpha p \tag{2}$$

$$mM[\text{BHCO}_3]=[\text{CO}_2]-0.0591\alpha p \tag{3}$$

$$p=\frac{[\text{CO}_2]}{0.0591\alpha(10^{\text{pH}-\text{pK}'}+1)} \tag{4}$$

the carbonic acid content of the larval blood as drawn is 0.99 millimole per liter, the bicarbonate content is 3.52 millimoles per liter, and the carbon dioxide tension ($p$) is 24.2 mm. of mercury.

The late instars of the southern armyworm have been found by Richardson [4] to be very resistant to nicotine. Sixth instars were therefore kept for 24 hours at room temperature in an atmosphere saturated with nicotine. The larvae, still alive although motionless, were then carefully washed with distilled water, the blood was removed in the usual manner, and its pH determined. Although nicotine is a strong base, having a dissociation constant of $9\times10^{-7}$, the pH of the blood was still its normal 6.65. Efforts were made to determine the amount of nicotine in the blood, but they were not entirely successful. The blood was made slightly alkaline with sodium hydroxide and distilled. The distillate had a distinct odor of nicotine, but the color obtained after treatment with Markwood's (*12*) reagents could not be matched directly with that of a nicotine standard. Since ammonia interferes with this test, the difficulty was probably due to the presence of ammonia, which was formed during the distillation from the urea and uric acid normally present in the blood. A match in colors was finally obtained by use of a Wratten 8 No. 58 filter, but too much faith cannot be placed in the quantitative nature of the result. By this method the blood contained 7.15 mg. of nicotine alkaloid per 100 ml.

## DISCUSSION

Since most metabolic waste products of animal origin are acid, one would expect the blood to be buffered mainly on the acid side. In the bloods that have been studied this has been found to be true.

The buffering power of human blood is due mainly to the alkaline salts of the proteins, hemoglobin being the most efficient, and to the bicarbonates. For the neutralization of volatile acids such as carbonic, hemoglobin is the most efficient because of its buffering action. For the neutralization of nonvolatile acids such as β-hydroxybutyric the bicarbonates are effective, not because of a true buffer action, but because of the efficiency of the lungs in removing the carbon dioxide that is liberated.

Since the buffer capacity is dependent on the concentration of the buffer material, the inorganic phosphates play only a minor role in the buffer system of human blood. Only 3 to 4 mg. of inorganic phosphorus is present per 100 ml. of blood, corresponding to about a 0.001-molar solution, and it has been found that normally blood can neutralize about 28 ml. of normal acid per liter within the physiological range.

At its average normal pH of 6.65 the blood of the southern armyworm has a buffer capacity of 0.022, which is approximately equivalent to a 0.038-molar solution and compares with the buffer capacity of 0.0228 for human blood at pH 7.4, as reported by Van Slyke (*17*).

In the insects whose buffer systems have been studied hemoglobin and hemocyanin do not occur; the buffering power of the blood is therefore due to an entirely different system from that of man and the marine invertebrates. The carbon dioxide content of human blood is between 45 and 60 volumes percent, about 95 percent of

[4] Unpublished manuscript

which is combined as bicarbonate. In insects the total carbon dioxide content is much lower, the blood of the southern armyworm containing only about 10 volumes percent, which corresponds to approximately 4.5 millimoles per liter, 3.52 millimoles, or 78 percent, of which is in the form of bicarbonate. Because of the small amount present it seems unlikely that bicarbonates are as active as buffers as they are in the higher animals. They do probably play some part, however. In the honeybee, where the average carbon dioxide content of the blood was found by Bishop (*3*) to be 29.2 volumes percent, the bicarbonates may play a more important role.

That compounds other than the salts of the weak acids are responsible for a considerable part of the buffering power of the blood of the insect is apparent from a consideration of the figures obtained by chemical analysis (*1*). Of the acids whose salts might act as buffers, inorganic phosphorus was 0.0056 molar, uric acid 0.00088 molar, and bicarbonates 0.00352 molar, a total equal to about a 0.01-molar solution. It must be noted, however, that the most effective buffer range of the salts of these acids is very near the normal pH of the insect blood. Van Slyke (*17*) has shown that the maximum buffer effect is exerted when $pH=pK'$. For the phosphates, urates, and bicarbonates these points are 6.8, 5.64, and 6.1, respectively.

It has been shown (*14, p. 892*), however, that, in addition to the inorganic phosphates, certain phosphorus-containing organic compounds also act as buffers in human blood. It is therefore probable that at least part of the organic phosphorus in armyworm blood is likewise available. This would, of course, reduce the buffering power unaccounted for, because the larval blood contains about 105 mg. of organic phosphorus per 100 ml., or about 0.034 mole per liter.

In human blood the content of the amino acids is so low that their slight buffering effect is exerted far from the neutral point. In insect blood, however, the amino acids are present in large quantities; the southern armyworm larval blood, for instance, contains 2.352 gm. of amino nitrogen per liter, which corresponds to about a 0.16-molar solution of amino acids. It is probable, therefore, that salts of amino acids and salts of proteins also play a considerable part in the buffering system of the insect's blood.

It is probable that in insects diffusion will account for both the intake of oxygen and the elimination of carbon dioxide. The insect possesses a circulatory system by which the hemolymph is kept in circulation, but so far as is known no experiments have been made that would show a difference in composition at any stage of the circulatory process. In man the reaction of the blood is dependent on the $BHCO_3/H_2CO_3$ ratio, which in turn is determined by the ratio of carbon dioxide production to elimination, while the $BHCO_3$ concentration depends on the amount of alkali in the body that is not combined with acids other than carbonic. The alkalinity of the blood as shown by the pH is a measure of the efficiency of the system, a low pH being direct evidence of the impairment of efficiency. It also seems probable that the low pH of insects' blood, being usually far on the acid side, is due, in some part at least, to inefficiency in the carbon dioxide excretory mechanism.

SUMMARY

The buffer capacity of the blood of sixth instars of the southern armyworm (*Prodenia eridania* (Cram.)) has been determined from titration curves. This value at pH 6.65, the normal for the blood, is 0.022.

The total carbon dioxide content of freshly drawn blood is 10.03 volumes percent, corresponding to 4.51 millimoles per liter. Of this, 3.52 millimoles is combined as bicarbonate and 0.99 millimole as carbonic acid.

Blood from larvae kept in an atmosphere saturated with nicotine for 24 hours did not change in pH, although the blood had absorbed considerable nicotine.

The buffering power of the blood is probably due to the presence of bicarbonates, phosphates, urates, and the salts of the proteins. The phosphorus-containing organic compounds and the amino acids also probably play an important role in the buffer system.

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